

A New Curriculum for the Schools ?

ONE of the principal preoccupations in British education is with the interface between secondary schools and later life, and particularly that between schools and the universities. The schools (or many of them) have traditionally thought of the sixth form, occupied chiefly by young people aged between sixteen and nineteen, as the limbo in which brighter students prepare to compete for the increasingly cut-throat competition for a place at a university, and in which lesser lights acquire the qualifications in the A-level examinations needed for some vocations, teaching for example. The result is that the sixth form is pedagogically both the best and the worst of British school education. It is a source of wonder that, in some schools, a handful of dedicated teachers can provide their star students with a large part of the curriculum they are likely to encounter when eventually they find their way to a university. But the sixth form curriculum is also a narrowing experience for these same students, who find themselves forced to specialize before their general education is complete. In the past few years, the stability of this system has in any case been undermined by the increasingly catholic interests of the students staying on into the sixth form, increasing numbers of whom are more concerned with vocational training of various kinds than with the academic rat-race.

This is why the Schools Council, chiefly responsible in Britain for innovation in the school curriculum, and the Standing Conference in University Entrance, the universities' watchdog of what are called "standards", have been labouring for the past four years on a formula to regulate the sixth form curriculum in such a manner as to cater for the increasingly diverse interests of students in the schools and the persistently rigorous expectations of university teachers, modified only to a small extent by their suspicion that it cannot be good, in some sense of that word, that all students over sixteen should be put through a specialized curriculum in order that the universities themselves should be relieved of what is ultimately their inescapable responsibility of deciding which students to accept. The outcome is a report from the Schools Council's Second Sixth Form Working Party under Dr E. W. H. Briault, and a companion document from the Joint Sixth Form Working Party under Dr C. C. Butler (Schools Council Working Papers 46 and 47). Mercifully, both documents say the same thing in different words. They agree that there should be a sixth form curriculum which entails that students completing five years of high school should follow a course made up of five distinct topics of study, and that those with academic inclinations should thereafter follow two of these topics in greater detail. Implicitly, the Schools Council committee report is a promise that secondary school teachers in Britain will accept this formula. Dr Butler's committee, by endorsing the same formula in many more words, implies a similar promise by the universities. It remains to be seen whether these promises can be met, but there is to be a period, no doubt lengthy, of feasibility trials which will allow both

partners to the present muddle about secondary education to declare themselves.

The first thing to be said is that the new proposals are an improvement. The idea that all entrants to the sixth form should follow a course of studies which has five topics and not three will, if found acceptable by the schools, be a step towards a liberal education. Unhappily, the new arrangements will not necessarily mean that those who wish in due course to become specialists of one kind or another will be required, during this initial period, to read widely, which implies that schools will in future, as in the past, be able to train specialists if they wish. Similarly, dedicated teachers and single-minded students will, if they choose, be able to sacrifice general education on the altar of supposed profundity. And so far as customers are concerned, there remains the danger that the new apparatus of school examinations will be used by the universities not merely as a means of telling which are the potential academics among school leavers but also, as in the past, as a device for selecting as university students only those who have already demonstrated that they can survive a university course as pastiched at school. In other words, quite apart from the objection that the new system will not specifically forbid the degree of specialization at school which has been traditional, there is a danger that it will more conspicuously drive a wedge, at the beginning of the sixth form, between those with academic inclinations and those with more general interests. Only the most fierce commitment to the cause of general education, in the schools as well as in the universities, will ensure that events take a different course. It is to be hoped that those with an interest in the feasibility trials are not exclusively those who seek to use the new system as a way of implementing courses of specialized study comparable in most ways with the system of the past several decades. The danger is that by a kind of pedagogical Gresham's Law, the bad currency of pointless, premature and unnecessary specialization will drive out the currencies of education in the round.

The second thing to say is that the new proposals, however they may be an improvement on present arrangements, are but an awkward device for avoiding several more fundamental questions about the relationship between school and university education in Britain. Both reports say explicitly that it is for universities to decide how students should be selected, but the British system is already shot through with the tradition that students should be selected for academic work in specialist departments at universities on the basis of their showing in similar studies at school. Is it not high time that British universities followed other university systems in asking whether the criterion of choice should be some more general test of a young person's academic potential? After all, less formally structured educational systems like the American, and more formally structured systems like the French, manage to base the selection of university students on a measure of fitness in general education.



Another unspoken and unanswered question is whether it is proper that the transition between school and university in Britain should take place, as at present, at eighteen or nineteen. Might it not be more economical as well as wiser and more suited to the needs of young people if what is called the sixth form were for most intending university students an experience lasting one or two years, not two or three, and if university courses were simultaneously lengthened from three to four? Most probably the answer would be that Mrs Margaret Thatcher, like her predecessors and probably her successors, would not sanction such a change. But there is no objective reason to think it would necessarily be more expensive while, educationally, it would almost certainly be more prudent. It may be that the best is the enemy of the good, which is why it may be wise that British education should now settle for the formula which the Schools Council and the Standing Conference on University Entrance have devised. But there is very little doubt that the proposed arrangements will pre-empt a more radical change for the best part of a decade.

France Should Not Test

THE government of France is no doubt righteously indignant that its plans for testing nuclear weapons in the Pacific have run into fierce opposition, not merely from the governments of New Zealand and Australia but from French citizens as well. For, the government of France may ask, did not the Soviet Union, the United States and the United Kingdom take advantage of testing in the atmosphere to develop their existing armouries of nuclear weapons? Does not the People's Republic of China still assume the right not to comply with the terms of the Partial Test-Ban Treaty, now nearly a decade old? What cause is there for asking that France should voluntarily forgo privileges which other nations have assumed.

The simple truth is that the testing of nuclear weapons in the atmosphere, which dates back to 1945, is a dangerous procedure whose dangers were not recognized until the mid-1950s and not acknowledged by the governments concerned until the early 1960s. But there is no doubt that if the military testing programmes of the superpowers had been begun at a time when the hazards of radioactive fallout were as widely understood as they are today, the protests would have been much more vociferous and the programmes more quickly brought to a halt. In retrospect, the existing nuclear powers may cynically be said to have been lucky that they began to develop their weapons at a time when ignorance of the consequences of radioactive fallout was widespread. (It is also, of course, fair to say that both the United States and the British governments of the 1950s did reprehensibly do much to minimize or even conceal the dangers of fallout, while the Soviet government has never openly admitted that these dangers are real.) But none of this can justify the way in which, a decade after the Partial Test-Ban Treaty, the French government now pretends that it too should enjoy the military convenience of a state of ignorance long since dispelled. The truth is that even if the series of tests now proposed by the French is numerically modest and even if the total yield of the explosions is comparatively small, there is nevertheless no doubt that the testing programme

will add to the reservoir of radioactive material in the atmosphere. Since there is every reason to think that many of the consequences of fallout are linearly related to the dose, however small it may be, the governments of all countries, not merely the governments of those in the immediate vicinity, have a right to protest at what the French government now plans.

So how should the government of France respond? The simplest and most honourable course would be to abandon the testing programme. On the face of things, such an act would also imply to outsiders that the French government's *force de frappe* was less of a strategic reality than Mr Michel Debré has been saying. In reality, however, the French government would be well advised to admit that the *force de frappe* is no more than an illusion and that its best course would be to use the protests at the proposed testing programme in the Pacific as an excuse not merely for abandoning the tests but also as a starting point for serious discussions, within the framework of the European Community, about long-term strategic defence.

What about China? The Chinese have of course been testing nuclear weapons for the best part of a decade, but only in the past year have they entered into meaningful diplomatic relationships with the rest of the world and the United Nations in particular. The first need is to make sure that the Chinese in their new role are prepared to follow the rules of diplomacy, such as they are; the experience of the past few months in the United Nations and of the bilateral negotiations with Japan and Western powers in particular have suggested that the Chinese are prepared to toe the line. So is there any reason why they should not now be asked also to comply with the spirit of the Partial Test-Ban Treaty and to abandon their own tests? The French would be on weaker ground if it were clear that the Chinese were also under pressure.

100 Years Ago



A GENTLEMAN writes us that he was invited by the Royal Commissioners to act as a juror at the Vienna Exhibition, but was at the same time coolly told that our Philistine Government had placed no funds at the disposal of the Commissioners wherewith to defray the necessary expenses of those who are willing to devote their valuable time and experience to the service of their country. Our readers will not be surprised at this. Other Governments have discovered that even in the most commercial, as well as in the highest light, the encouragement of science "pays." The British Government, with five millions on the right side of their account, still regard science as a beggarly Lazarus, to whom, for mere shame's sake, they are compelled to throw an occasional crumb. As our correspondent says, poor little Switzerland has devoted two and a half times the pittance our Government have allowed to defray the expenses of the Vienna Commission; while the amount expended by Austria in their department of former exhibitions was at least four times as much as we have devoted to theirs.

From Nature, 8, 54, May 15, 1873.

OLD WORLD

Fusion Research at Culham Expands

OUTLINE design studies on a fusion reactor are under way at the Culham laboratory of the United Kingdom Atomic Energy Authority. Work in the past five years has led to a much more optimistic assessment of the chances of fusion being used to provide electrical power, and support for the AEA's fusion work is to be increased from £3.9 million this year to £5 million a year by 1975-76. This reverses the decision taken in 1968 by the Labour Government and Mr Anthony Wedgwood Benn to halve the support for fusion research.

The decision to increase support follows the report by Sir Harrie Massey to Mr Tom Boardman, Minister for Industry, in which the AEA's fusion work was examined.

Dr R. S. Pease, Director of Culham, said last week that developments in high temperature plasma physics, in the understanding of toroidal magnetic field systems, particularly toroidal pinches, and in methods of suppressing the rapid diffusion (Bohm diffusion) that leads to poor thermal insulation, have all made the practical development of a fusion reactor a distinct possibility.

The enlarged programme has also been made possible by cooperation with Euratom. The—at any rate partial—solution to Euratom's future that was agreed on February 5 included an increased fusion research programme. Although this decision has still to be ratified, the Culham fusion programme will be jointly managed by the UKAEA and Euratom as part of a Euratom programme that will be carried out chiefly in national laboratories and which will be running at between £20 million and £25 million a year. Research at Culham will constitute about a quarter of the total, Dr Pease said, and about £1.25 million of the money spent at Culham will come from Euratom. The remainder will be provided by the UKAEA.

Dr Pease was also at pains to stress the importance of cooperation with the United States and the Soviet Union, and he cited the studies carried out in 1969 with the Soviet Union on Tokamak T-3 which has provided some of the best results yet with these experimental fusion assemblies.

The eventual aim of the enlarged programme at Culham will be to produce a plasma with a temperature of 100 million K and a density of 10^{14} cm⁻³ with a confinement time of over a second. Culham will try and reach this goal with continued effort on confining magnetic

fields within toroidal magnets. At present Culham has three new experimental assemblies in operation in order to achieve this goal. First, there is a superconducting levitron; second, a high beta toroidal experiment (HBTX); and third, a CLEO Tokamak/stellarator experiment. The laboratory also plans to build further stellarator and Tokamak apparatus in the near future.

Each one of these pieces of equipment enables different facets of fusion to be examined. In the Levitron, for example, long confinement times can be studied—times of up to 1/10 sec have been obtained with this apparatus but the density of the plasma has been low. In the HBTX there is a dense plasma but unfortunately a lower containment time, while in the CLEO Tokamak apparatus discharges lasting 1/10 sec have been obtained with poorly conducting walls (such as would be obtained in a reactor) but when a combination of optimum density and good temperatures is obtained the confinement time drops to 1/100 sec.



Dr R. S. Pease

The next step according to Dr Pease is the construction of a large apparatus in which a high density, high temperatures and a long containment time can be simultaneously maintained. The apparatus will be toroidal and probably a tokamak. Culham is at present actively discussing with Euratom where this apparatus should be constructed. But wherever that might be it is planned to have it in operation by 1978.

If the problems of maintaining a plasma at high temperatures and high

density for long times can be beaten then the next step is to develop a fusion reactor that can produce electricity commercially. This is a long way ahead although Dr Pease feels that when the large apparatus is built there will be a chance, however slight, that fusion power will be a reality before the end of the century. It remains to be seen whether this is more of a hope than an expectation.

EMBO

New Secretary

DR JOHN TOOZE is to be the Executive Secretary of the European Molecular Biology Organization. The decision, taken by the EMBO council last week, will mean that Dr Tooze will relinquish his position as research administrator at the Imperial Cancer Research Fund in London. Dr Tooze has been a regular contributor to *Nature* for the past seven years, and was deputy editor in 1968 and 1969.

Dr Tooze takes over the job from Dr Raymond Appleyard who is himself taking up another job with the European Commission in Brussels. Dr Appleyard, who was director and head of the biology programme in the Industrial, Technological and Scientific Affairs division, is now the Director General in the Dissemination of Information division.

The agreement among ten of the fourteen EMBO countries to build a European Molecular Biology laboratory in Heidelberg is to be signed later this week in Geneva. Before building can start the agreement will have to be ratified by the respective governments, a process which could take as long as a year. Although EMBO and the proposed laboratory are not directly linked, the new Executive Secretary will set up office in Heidelberg, but this will not happen for about 18 months. In the meantime EMBO will continue to be managed from Brussels.

CRIMOND TERMINAL

Loch Reprieved

TOTAL Oil Marine and the British Gas Corporation have put their plans to build a gas processing plant above the Loch of Strathbeg near Aberdeen into abeyance. The purchase of an alternative site at St Fergus, three and a half miles south of the loch, is being arranged

and the plant may well be built there.

The change of plan became apparent when Mr F. E. Dean, the Gas Corporation's Chief Technical Liaison Officer, wrote to Aberdeen County Council informing it that the alternative planning application was being prepared. The chief reason for the change is said to be that the Ministry of Defence, which already has planning permission to build a radio station on Crimond airfield, above the loch, is still eager to use the site. A Gas Corporation spokesman added this week, however, that the protests from conservationists in the area who were worried that the plant might seriously damage the loch, which the Nature Conservancy classifies as being of grade one scientific importance (see *Nature*, **242**, 93; 1973), did have some effect.

The St Fergus site is much less valuable than the Loch of Strathbeg which is Britain's largest dune slack pool. The slack pools, dunes and bird-life at Blackwater, just north of St Fergus, where it is thought the plant will be sited, are far less likely to be disturbed by the station than at the Strathbeg site.

The Gas Corporation was at pains to point out this week that planning application for Crimond has merely gone into abeyance, and if the Blackwater site proves impossible, Crimond may still be the chosen spot. It is unlikely, however, that Total and the Gas Corporation would get the site without a planning inquiry if the Ministry of Defence seriously wants it. Work was due to start on the ministry's radio station this year, with completion planned for 1975, but the plan to put a gas processing plant on Crimond may have held work up.

BRITISH MUSEUM

Chaotic Jubilee

THE British Museum Research Laboratory is celebrating its Golden Jubilee with an exhibition which opened on May 4. At the entrance to the exhibition blowups of several historic press cuttings are displayed in a sepia coloured anteroom simulating the conditions under which the first workers in the laboratory laboured. Among these cuttings is one from the *Daily Herald* of December 31, 1931:

"I am certain that Dr Alexander Scott (FRS) could put Humpty Dumpty together again. That would be a small job for the head of the laboratory which cleans and restores the priceless exhibits of the British Museum."

So begins the cutting. And perhaps Dr Scott's sure touch would have helped in the preparation of this exhibition, for at the press showing on May 3 the display was still being put together, and

seemed to be in about as many pieces as Humpty Dumpty after his fall. That makes it difficult to assess the merits of the exhibition, housed within a double dome inflated by air pressure, in the former ethnography gallery. Undoubtedly the show will be worth a visit in calmer conditions, when it will be possible to see if the rainbow coloured lighting and technological exhibitionism of the surroundings add to, or detract from, the exhibits, which will include the use of radiography, metallography, X-ray diffraction analysis, atomic absorption analysis and both carbon-14 and thermoluminescence dating.

The overall appeal of the exhibition seems likely to be visual rather than scientific. One already complete exhibit, for example, shows a glass vase which appears pea soup green in reflected light and magenta in transmitted light. This effect has been duplicated at the laboratory by adding sundry traces of metallic oxides to glass—but this seems very much window dressing compared with the real work of the laboratory.

It is, however, obvious that the staff of the laboratory has achieved much under difficult conditions. Not only scientific problems but bureaucratic difficulties have confronted them. In another cutting displayed at the show, quoting from the *Birmingham Post* sometime in 1932, we learn that "The laws and regulations under which museum authorities work are strict. As a precaution against fire and explosion there is no gas on the premises. Some years ago a room was set apart as a laboratory, and a supply of gas became essential, but the regulations could not be broken. The difficulty was overcome by carrying a pipe to the outside of the laboratory window, and then continuing the supply to the desired position by means of a rubber tube that could be disconnected every night".

Also, "No exhibit can be taken away for treatment. Once an exhibit has been accepted by the trustees an Act of Parliament would be necessary to permit its removal from the premises. The laboratory in Russell Square . . . is made technically part of the Museum by a connecting corridor across the grounds. Every article in need of treatment . . . has to be carried that way so that there shall be no direct breach of the rules." And seemingly it still requires an Act of Parliament to remove articles from the Museum, for example when exhibits are displayed abroad.

Although when the laboratory was first set up in 1922 *Nature* commented that "National laboratories for the cleaning, restoration, and preservation of antiquities have for some years existed at Berlin, Copenhagen and Stockholm" (109, 119; 1922), it is today the oldest museum laboratory in exist-

ence. So, having started perhaps somewhat ingloriously, primarily concerned in the first years with the restoration of objects which had been damaged when stored in a section of the Underground during the First World War, the staff have now thoroughly earned their right to a Jubilee junket.

PESTICIDES

Too Much Concern

CONCERN about the use of pesticides has sometimes been due to misinterpretation or improper extrapolation of scientific data, according to a World Health Organization expert committee on insecticides (*Safe Use of Pesticides*, Report No. 513, WHO, Geneva, 1973). This has caused some administrations to impose restrictions on the use of certain pesticides which cannot be justified on the basis of present scientific knowledge, says the committee.

The nub of the report is that the advantages of the use of pesticides, in particular DDT, in controlling diseases outweigh their disadvantages. The committee warns that the withdrawal of DDT from areas which in the past have been ravaged with malaria could well see the return of the disease.

But the committee accepts that there is insufficient evidence available on the effects of DDT on man. It recommends that comprehensive metabolic studies be carried out of the effect of the disease to complement studies on the carcinogenicity of the pesticide. The committee points out, however, that even though the first reports that DDT might cause tumours in rats appeared in 1947, there has been no adverse effect observed in a small group of men who have been exposed to heavy doses and who have been under survey for the past twenty years.

Education is the answer to the hysteria caused in recent years by the supposed effects of pesticides according to the committee. The report says that even in developed countries, people need to be introduced to the basic principles of toxicology "in order to dispel the excessive concern over hypothetical but unproven effects of pesticides and to ensure a more realistic appreciation of hazards".

DDT is not the only topic covered in the report and the committee also makes a plea for the introduction of less toxic mercury compounds than those at present used as seed dressings and fungicides. The alkyl mercury compounds, the committee points out, accumulate in the nervous system giving rise to irreversible damage. So, "in the long view, it may be unwise to use any mercury compounds and certain other heavy metal compounds as pesticides".

NEW WORLD

Functional Organization Makes Trouble

by our Washington Correspondent

A SWEEPING reorganization of the federal government's health services has been set in train by Caspar W. Weinberger, the new Secretary of Health, Education and Welfare (HEW). Tighter management control is the watchword, and although some reorganization of HEW's sprawling bureaucracy takes place nearly every year, this particular round of executive musical chairs is of more than passing interest to the scientific community. The chief losers in the shuffle are mental health programmes in general and the National Institute of Mental Health (NIMH) in particular.

The nub of the plan is that the Health Services and Mental Health Administration should be dismantled, and that all the health programmes of the federal government should be reorganized into five separate agencies, the heads of which will report directly to Dr Charles C. Edwards, the new Assistant Secretary for Health. NIMH is being transferred almost lock, stock and barrel back into the National Institutes of Health, the agency which housed it until 1967 when NIMH was separated from the other biomedical research institutes and promoted to bureau status.

The five health agencies to be established by the reorganization are as follows:

- The Food and Drug Administration, which will be unchanged.
- The Center for Disease Control in Atlanta, Georgia, which will also be essentially unchanged but which will take on the National Institute for Occupational Safety.
- The National Institutes of Health, which will gain NIMH, but lose the Bureau of Health Manpower Education.
- A new Health Services Administration, which will be concerned with health care delivery.
- A new Health Resources Administration, which will be concerned with health service data gathering and demonstration programmes, and which will also take over the Bureau of Health Manpower Education from NIH.

Harold O. Buzzell, former Deputy Manpower Administrator for the Department of Labor and a management consultant who once worked for Edwards's former employers, Booz, Allen and Hamilton, has been appointed to implement the reorganization and eventually to direct the Health Services Administration.

The objectives of the shuffle are clear:

to reorganize the health bureaucracy along more functional lines and to put the Assistant Secretary of Health in firm control of the federal government's health programmes. In the past, the assistant secretary's lines of control have been rather obscure, and he has often been bypassed on crucial decisions.

What all this means for biomedical research and the National Institutes of Health is a matter of debate, but a clear indication of the way the wind is blowing was given by Mr Weinberger during a briefing for a few reporters before the plan was announced. He was quoted as saying that the reorganization will help to "redress the balance" in research funding. "There has been too much emphasis on investigator-initiated research and not enough on directed research", he is reported to have said, and suggested that more research fund-

ing should be spent on attacking specific diseases such as cancer, and less on basic research.

Concentration on research likely to produce results in the short term at the expense of longer-term studies has become the general trend in research spending across the board in the United States in the past few years, but if Mr Weinberger had intended to antagonize much of the biomedical community, he could hardly have chosen his words more carefully. Already the cancer plan, which has not yet been published, has been bitterly attacked for stifling scientific creativity, and budget cuts which have reduced general research support in every institute in NIH have been attacked for jeopardizing the long-term future of biomedical research in the United States.

In organizational terms, the reorgani-

MARS

Targets for Viking

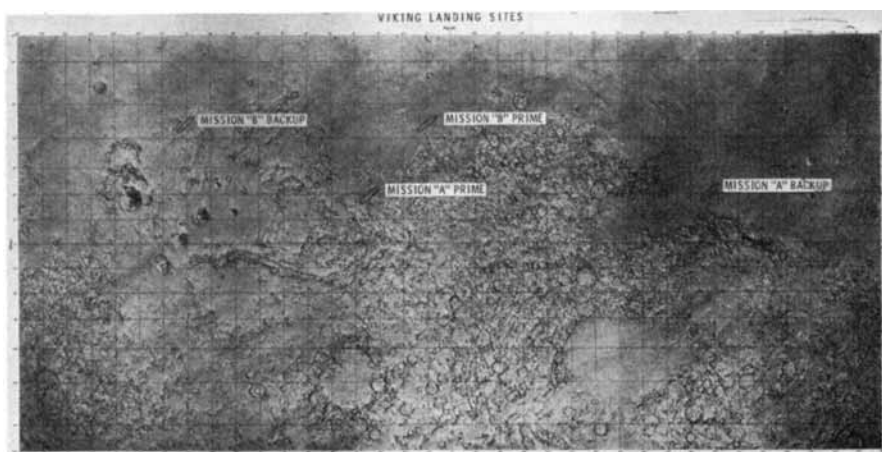
POSSIBLE landing sites for the Viking mission to Mars in 1976 have finally been chosen by NASA. After a year's intensive study of some 22 possible sites, NASA has chosen Chryse (19.5° N, 34° W) as the prime target for the first lander and Cydonia in the Mare Acidalius (44.3° N, 10° W), as the target for the second lander.

Chryse is situated at the northeast end of the 3,000-mile long, 20,000 feet deep rift valley discovered by Mariner 9, in an area which resembles a drainage basin, and it is expected to contain a variety of deposits of surface material. The first lander will visit the site in June 1976 if all goes well. The second landing site, which will receive another lander a month later, lies at the edge of the southernmost regions of the north

polar hood — a hazy area which may carry moisture. Chryse is thus more interesting geologically, while Cydonia offers the best prospects of any life.

Both sites also lie well below the mean surface of the planet, which is an advantage in two respects. First, the denser atmosphere will provide more efficient braking for the parachutes, and second, the high atmospheric pressure adds to the chances of finding water. Backup sites for each lander are at the same latitudes as the prime sites.

A special advantage of the two prime sites is that both landers will be in communication range of both orbiters, which could enable one orbiter to be released from synchronous orbit for low-altitude study of other regions, while the second orbiter relays information back to Earth from both landers. The sites are tentative, and can be changed by retargeting the spacecraft up to 10 days before encounter.



zation plan has some interesting implications for NIH. First, after six years of independence, NIMH is being returned to the NIH fold. As far as NIMH's biomedical research is concerned, the transfer will probably not make much difference because it never really left the NIH campus, and there has continued to be considerable integration of facilities and programmes. Dr John Eberhart, Assistant NIMH Director for Mental Health Research, has continued to attend meetings of NIH programme directors, for example, and the NIH Division of Research Grants has provided many of the same services for NIMH that it provides for other institutes of NIH.

But the transfer does bring into the NIH many service activities which have not previously been its concern (although the cancer bill has recently given NIH authority to run cancer control programmes). Many such health services are connected with NIMH's drug abuse activities, which are being kept in the institute more because of legal difficulties than anything else. The Administration has been toying with the idea of either splitting them off into a separate addiction agency or transferring them to the Center for Disease Control, but because they were originally consigned to NIMH by act of Congress, they could only be split off with Congressional approval.

As for NIH's Bureau of Health Manpower Education, the Administration's proposed budget cuts and the non-renewal of legislation dealing with manpower for health and allied professions have severely reduced its activities and effectively separated its programmes from other NIH activities. Although the events which have led to such a situation have been painfully felt and bitterly attacked in the medical schools, they have removed much of the original justification for placing the bureau under NIH control.

The transfer of NIMH back to NIH has been called a downgrading of the federal government's mental health programmes. That may be so, but for NIMH the writing was on the wall in January, when President Nixon's budget was unveiled. The budget proposed phasing out federal funding of community mental health centres (CMHCs), a programme launched by President Kennedy in 1963 as a bold new approach to caring for the mentally ill. The CMHC programme has resulted in the setting up of about 400 centres providing mental health care on a community basis, funded partly by NIMH and partly by state and local governments. Ironically, the Administration has proposed that federal support should be withdrawn because the centres are so successful—the argument is that the states will pick up funding with money from general revenue sharing.

Sakharov Honoured

THE National Academy of Sciences has elected Dr Andrei D. Sakharov, the dissident Soviet nuclear physicist, as a foreign associate. Sakharov, who played a leading role in development of the H-bomb in the Soviet Union and who has recently been in trouble with Soviet authorities for his championship of civil liberties, is one of only 12 scientists to be elected as foreign associates of the academy this year. Dr Philip Handler, NAS President, said last week that Sakharov's election was "by no means for political reasons"—his nomination came from the academy's physics section for original scientific research. He is, in fact, one of three Soviet scientists to be elected as foreign associates of the academy last week, the others being Vladimir Engelhardt, Institute of Molecular Biology, and I. S. Shlovsky, Institute of Space Research.

Other foreign associates elected were Sune Bergstrom, Karolinska Institutet, Stockholm; Albert Eschenmoser, Technische Hochschule, Zurich; Dennis Gabor, Imperial College, London; David Glass, London School of Economics; Jean Goguel, Ecole des Mines, Paris; Motoo Kimura, National Institute of Genetics, Japan; George Klein, Karolinska Institutet; Ben Roy Mottelson, Institute for Theoretical Physics, Copenhagen; and V. Ramalingaswami, All-India Institute of Medical Sciences, New Delhi.

NAS/NAE

Options Kept Open

by our Washington Correspondent

THE annual meeting of the National Academy of Engineering last week passed a resolution requesting the NAE's governing council to prepare a plan for incorporating the academy as a non-profit corporation separate from the National Academy of Sciences. But several amendments to the resolution, proposed from the floor, have kept options open for the NAE and they should help to ensure that the two academies will be able to live in harmony whether or not their planned divorce is made final.

In short, the resolution authorizes (but does not direct) the council to seek approval at its annual meeting in October of its plan to establish the NAE as a corporation, but it also suggests that

negotiations should be continued with the National Academy of Sciences, that a Congressional Charter should be prepared and that the members should be given at least two options for action before the next annual meeting. The chances are that the NAE will set up shop independently of the NAS, but NAE officials emphasized after the meeting that whatever the final outcome of the breakaway movement, they hope that the two academies will continue to work together on problems and studies of mutual interest.

The basis of the long-standing disagreement between the two academies is the management of the National Research Council, their joint operating arm. Governance of the NRC is vested solely in the hands of the Council of the NAS, and NAE officials want it to have a tighter management structure (see *Nature*, 243, 5; 1973). The NAE, which was set up in 1964 and has since been operating under the charter of the NAS, also wants to be more visible in the engineering community.

Shockley Again

FOR the past seven years, Dr William Shockley, co-inventor of the transistor, has been trying to persuade the National Academy of Sciences to examine the evidence for his belief that genetic factors make blacks less intelligent than whites. He failed again last week when the annual meeting turned down his familiar resolution calling for a committee to study the matter, but this year the meeting passed a substitute resolution which may help to blunt criticism from Shockley's supporters that the academy is treating him like a latter-day Galileo.

Offered by the academy's governing council, the substitute resolution acknowledged Shockley's efforts to bring his studies to the academy's attention, and encouraged "him and others engaged in such efforts to follow the normal scientific procedures of publication so that their results may be subjected to the usual peer review and accorded the scientific impact afforded by such publication". A preamble to the resolution said that Shockley's previous attempts had been voted down without debate because "only a small minority of the membership has any expertise in the field and . . . many of those with expertise doubt that the field has progressed to the point at which a definitive conclusion is warranted".

NEWS AND VIEWS

Unravelling the Eukaryotic Chromosome

RECONCILING the visible structure of the eukaryotic chromosome with its molecular organization has for some time seemed to be an almost insuperable obstacle in preventing any real advance in understanding the control of gene activity at the molecular level. The chromosome has appeared more complex than the sum of its parts and the environment of the gene has remained virtually undefined. At the visible level, chromosomes contain two types of region—euchromatin, which has a diffuse structure and is active in the synthesis of RNA, and heterochromatin which appears less compact and is inactive in gene expression. Early attempts to use these two types of structure by finding differences between them which might throw some light on the structure of the chromosome all failed. But evidence that regions of the chromosome may be distinguished by their responses to certain dyes has been accumulating in recent months, and Comings and his colleagues now report in *Experimental Cell Research* (77, 469; 1973) that these experiments may reflect the uneven distribution along the chromosome of non-histone proteins and not, as had at first been thought, differences in sequences of DNA.

The first breakthrough in defining the structures of heterochromatin and euchromatin was provided by the demonstration that the heterochromatin at the centromeres of eukaryotic chromosomes is rich in sequences of satellite DNA, the component of the genome which consists of a short basic unit repeated many times in related but non-identical copies. The cytological hybridization used for these experiments showed that the centromeric heterochromatin is distinct in two respects; it hybridizes with satellite DNA sequences and it stains very deeply with Giemsa stain after the hybridization. When Arrhigi and Hsu and Yunis *et al.* showed that staining takes place in such heterochromatin whenever the chromosomes are treated with alkali and then incubated in a saline solution at 60° C for some hours—part of the protocol followed in cytological hybridization—it was thought that the unusual properties of the DNA might explain the deep staining.

An equally important discovery about chromosome structure within the euchromatin or the arms was the observation of Caspersson and his colleagues that treatment with quinacrine causes each chromosome of the human complement to fluoresce intensely in localized regions. In fact, the entire chromosome complement may be identified by the characteristic fluorescent banding patterns of each chromosome. A connexion between Giemsa staining and quinacrine fluorescence was then suggested by the work of Evans and his colleagues, who showed that the same banding patterns are produced by a very gentle treatment of the chromosomes with saline solution followed by staining with Giemsa. These techniques, which are now coming into widespread use, have been named C banding (alkaline treatment, incubation in saline solution at high temperature, Giemsa staining), Q banding (direct application of quinacrine) and G banding (incubation for 1 h in saline solution, staining with Giemsa).

An early hint that the preferential renaturation of

repeated DNA sequences might be irrelevant to banding was provided by Stockert and Lisanti (*Chromosoma*, 37, 117; 1972), who found that the same C banding pattern is obtained no matter whether the DNA is single stranded or double stranded. By using the reagent acridine orange, which fluoresces orange with single strands of DNA and green with duplex strands, they demonstrated that the centromeric regions, as expected, renature the most rapidly after denaturation; but renaturation of all regions is complete within a few minutes. Comings *et al.* have now excluded another possible explanation—that the DNA is more concentrated within the centromeric heterochromatin and thus stains more deeply—by measuring the content of DNA along the chromosome with feulgen stain. The packing of DNA in centromeric heterochromatin seems if anything to be slightly less compact than in euchromatin. (They therefore suggest that the compression of DNA in heterochromatin may be relatively fixed and constant during the cell cycle; during interphase, when the euchromatin uncoils to a diffuse structure, heterochromatin therefore provides the most condensed regions but at mitosis when the euchromatin becomes compact also there is little difference between the structure of the two regions.)

The alkaline treatment which is used to denature DNA in the protocol for cytological hybridization seems to be the critical step in generating the intense C bands at the centromeres instead of the G bands along the chromosome arms. Comings *et al.* report that treatment with 0.07 M NaOH for 30 s removes 21% of the DNA from the chromosomes; and the incubation in saline overnight at 60° C which is the next step extracts a slightly greater amount. By the time the chromosomes are stained with Giemsa then, they have lost half of their total DNA; and the extracted sequences all derive from the arms rather than from the centromeres. The repetitive sequences of centromeric DNA must therefore be packaged in a structure much more resistant to extraction than that of the arms.

Is the preferential extraction of DNA sufficient to explain C banding? Because gentle treatment with trypsin or other proteolytic enzymes produces a G banding pattern in the arms, but more prolonged incubation abolishes the G bands and instead produces C bands, Comings *et al.* suggest that differences in the proteins associated with DNA may explain both C banding and G banding. Because extraction with acid does not produce a banding pattern, it seems likely that the non-histone proteins must be responsible for the definition of regions as bands or non-bands. According to this model, a gentle disruption of chromosome structure by short incubation in saline, treatment with trypsin and so forth, extracts proteins from the chromosome. Untreated chromosomes stain with Giemsa uniformly along the arms, but react less intensely at the centromeres. The extraction must therefore remove some proteins from the regions which cease to bind Giemsa, that is the non-bands, leaving Giemsa able to bind only to the bands retaining the proteins. More stringent treatment, either with saline at high temperature, or with trypsin, or with alkali, completely disrupts the

structure of the arms by extracting either their proteins or DNA; in either situation, the G bands and Q bands are abolished and the centromeres are converted in to a state in which they stain intensely instead of failing to react.

Three types of structure can therefore be defined in the chromosome by the banding techniques. Centromeres contain heterochromatin which is the least reactive region in G banding or Q banding but reacts intensely under the conditions of C banding. The arms of the chromosomes can be divided into bands which react with Giemsa and quinacrine and non-bands which fail to do so. One puzzle about the nature of the bands is that they react with quinacrine directly, but react with Giemsa only after pre-treatment has extracted some of the non-histone proteins, presumably from the non-banding regions. The similarity of the results produced by these two different treatments therefore argues that the bands may correspond to some important underlying structure of the chromosome and are distinct from the non-banding regions.

What is the nature of the material which forms bands? The centromeric heterochromatin contains sequences of highly repetitive DNA and in organisms where such sequences occupy extensive regions of chromosome, they too show C banding. This suggests that the repetitive satellite sequences differ in their chromosomal proteins from less repeated sequences; this provides the first evidence to show that the distribution of proteins in the chromosome may be segregated according to the function of the different regions. But the properties of the bands in the arms is less well defined. It is tempting to speculate that they too form heterochromatin; but this conclusion rests solely upon their staining properties—the traditional distinction of heterochromatin—because they have not yet been shown to have any of the other features characteristic of centromeric heterochromatin, such as genetic inactivity, late replication or repeated sequences of DNA. In fact, one of the most important conclusions drawn from the experiments of Comings *et al.* is that the evidence of previous experiments that repeated sequences are not present in the arms must be reconsidered; because these experiments used alkali, much of the DNA of the arms must have been extracted before the hybridization step, so that it gives little information about the sequences of the arms. The parallel between the C bands on the one hand, and the G bands and Q bands on the other, is therefore intriguing but incomplete; whether or not the bands are heterochromatin, however, they represent sequences of structural distinction which if correlated with specific functions (or lack of functions) may begin to help to unravel the structure of the chromosome.

From our Molecular Genetics Correspondent

Block Copolymers

THE rationale of materials science as a discipline lies in the stimuli that the student of one category of materials derives from the study of a quite different category. Such stimuli most commonly derive from studies of microstructure and the experimental factors that control it. For a long time now, students of metals and of ceramics have learned from each other in this way. Until quite recently, polymers have been a family apart, but as

progressively larger and more perfect polymer crystals have been grown, the whole metallurgical apparatus of crystal growth, grain structure and dislocation morphology has become increasingly relevant to polymers. Recent developments in the formation of block copolymers have now taken this slow rapprochement a stage further; from now on the polymer scientist will need to concern himself with the microstructural disposition of two phases.

A block copolymer is ideally one in which a substantial block of one homopolymer is joined to one or more blocks of a chemically distinct homopolymer. A particularly important example is the rubbery (SBS) copolymer consisting of a polybutadiene block flanked by two polystyrene blocks (Holden, Bishop and Legge, *J. Polymer Sci., C*, **26**, 37; 1969). For these novel materials to develop their excellent mechanical properties, the individual blocks need to have closely controlled, uniform lengths—that is, both the total and the partial molecular weight must be fixed. This has become feasible through the technique of “living” polymerization, originated by Szwarc (*Nature*, **178**, 1168; 1956). This very important method depends on a special (anionic) method of maintaining the polymerization reaction so that no accidents can happen to “kill” the growing end of a polymer chain and thus lead to a broad spectrum of chain lengths. By ensuring rapid initiation and then controlling the growth of the population of chains by varying the species and concentration of monomer to which the population is successively exposed, block copolymers of rigorously controlled total and constituent molecular weights ensue.

Szwarc, who works at the State University of New York at Syracuse, has now published the first extensive review of the synthesis and properties of block and graft copolymers made by the living copolymer technique (*Polymer Eng. Sci.*, **13**, 1; 1973); every teacher of materials science should read it. Whether or not he is interested in the chemical details of synthesis, he is bound to be stimulated by the account of the microstructure of these polymers and their closely related properties. As the styrene/butadiene ratio is altered, two-block copolymers of these monomers show a sequence of structures closely reminiscent of metallic eutectics. The minority phase forms a regular array of spheres; if its concentration is increased, the spheres give way to a hexagonal array of rods and these in turn to a lamellar morphology. Finally, matrix and disperse phase are inverted as the concentration of the original minority phase is further increased. One difference from metallic eutectics is the much finer scale; polymer eutectics typically have a periodicity of only 10–20 nm. Another difference is that the phases are probably not to be regarded as true phases at all—Szwarc dubs them “mesophases”—because individual polymer chains are long enough to pass through several “interphase” boundaries.

Microstructures of SBS rubbers, which are similar, have been assiduously studied at Manchester University by Lewis and Price, for example, *Laboratory Practice*, **19**, 599; 1970). They produced their polymers in the form of electron-transparent films by solvent evaporation, and found that the scale of the pseudo-eutectic structure is modified by a change in the rate of evaporation. One is irresistibly reminded of the modification of metallic eutectic microstructures by a change in the freezing rate. Lewis

ZOOS

Breeding Waterfowl in Captivity

THE indispensable *International Zoo Yearbook*, (vol. 13, Zoological Society of London, 1973, £8) this year has a special section on waterfowl in captivity. The young of the rare New Zealand brown teal (*Anas aucklandica chlorotis*) shown here were hatched for the first time in captivity at the Wildfowl Trust, Slimbridge. Both parents tend the young, an unusual feature in dabbling ducks.

and Price point out that the microstructure of a block copolymer has four levels of organization: primary chain structure; geometrical conformation of individual chains; microphase separation and disposition; and "grain structure". This last refers to the existence of regions in which the hexagonal array of polybutadiene microspheres lies on one "lattice", abutting on other regions or "grains" in which the lattice has a different orientation. Lewis and Price have even studied the arrangement of microspheres in the "grain boundaries", which are disturbed just like real grain boundaries in metals. It will certainly benefit the development of materials science that Lewis, a chemist, is now pursuing these studies in a metallurgy department.

Returning to Szwarc's review, a number of illustrations may be found of the advantages of a precise control of the chain structure of a block copolymer. (Note that the living polymer technique allows a polymer of a given gross composition to be produced in a variety of microstructures, corresponding to different block lengths, just as an alloy of fixed composition can be given different microstructures by varied heat-treatments.) The essential physical and chemical properties of the constituent blocks are preserved and not averaged out as in a random copolymer. Thus weak but permeable desalination membranes of cellulose acetate can be stiffened by making a block copolymer with polystyrene, which is more resistant to creep though not by itself permeable.

Again, microstructures with dispersed arrays of rods can be stabilized by dissolving a block copolymer made by Szwarc's method in a suitable, chemically distinct monomer and then rapidly polymerizing the monomer. The rod structure, already existing in the solution, is thus fixed in a matrix which now incorporates the second polymer, and the whole is a form of ultrafine fibre-reinforced polymer, which can be aligned by extrusion.

Szwarc suggests a number of other technologically desirable properties which could be achieved by special forms of highly regular block copolymers, and calls for intense studies in this field. The call is fully justified.

From our Materials Science
Correspondent

PROTEINS

Built to Last

from our Molecular Biology Correspondent
GIVEN the facts of hydrophobia in proteins, one would suppose that the lower limit of chain length to allow the formation of a stable globular conformation would be governed by the number of non-polar side chains needed to generate an "inside" from which water is excluded. So far the smallest proteins that can be categorically said to possess a unique, stable globular structure have fifty to sixty residues, and include the insulins ferredoxins, a family of venom neurotoxins and pancreatic trypsin inhibitor. This last is a remarkable specimen, for in spite of its small size, it exhibits a quite remarkable stability towards denaturants, such as guanidine hydrochloride, renatures from the

reduced state with unusual speed and efficiency, and can be kept in solution without loss of activity literally for years. Its sequence and structure are known; it is without histidine or tryptophan, but has three disulphide bonds, providing a rather high cross-link density by the standards of larger proteins. Some remarkable aspects of its stability are now revealed in proton magnetic resonance studies from two laboratories.

Masson and Wüthrich (*FEBS Lett.*, **31**, 114; 1973) have concentrated their attention on the downfield region of the spectrum, where, below the phenylalanine and tyrosine ring resonances, an extensive series of peaks, mostly corresponding to single protons, and many of them resolved into doublets, are apparent in freshly prepared D_2O solutions. These are evidently refractory exchangeable hydrogens, presumably of internal peptide groups, showing coupling of the amide proton pairs. Guanidine hydrochloride makes no impression on these resonances until the solution is heated, when, at a sufficiently high temperature, they vanish. Renaturation by diluting out a solution of the protein in dimethyl sulphoxide with D_2O regenerates the NMR spectrum characteristic of the native state, except for the downfield peaks. Now resonances from shielded exchangeable protons have been observed in fresh solutions of a few other proteins, but they disappear quickly. In pancreatic trypsin inhibitor, on the other hand, of the fifteen observed features of this nature, four diminish noticeably in intensity on a time scale of hours, another six after several days, but the remainder appear to be indefinitely stable. With sufficient effort and ingenuity it may be possible to make specific assignments in terms of Huber's high-resolution X-ray structure. Slow exchange prevails also at pH 0.7, and even in alkali at pH 11 and higher, three of the shielded protons persist. Exchange in the native protein can be provoked by heating, and occurs in minutes at 65° C, whereas no unfolding can be detected by any of the usual methods until the temperature reaches 85° C.

Much the same results have been obtained by Karplus, Snyder and Sykes (*Biochemistry*, **12**, 1323; 1973), who waited for four months for the downfield resonances to go away, and at the end of this time the greater number were still there. The usual view of protein conformation is that it allows for a certain measure of undulation or "breathing". By the standards of other globular proteins that of pancreatic trypsin inhibitor is evidently quite remarkably resistant to any such fluctuations, except at high temperature; that is to say, there is an energetic barrier to any kind of unfolding of quite

astronomical proportions.

In the alkaline range, the aromatic ring proton resonances of the tyrosine residues undergo shifts with the ionization of the phenolic groups. Thanks to the absence of other aromatic residues apart from phenylalanine, the spectrum in this region is relatively simple, and because the magnetic environments of the four tyrosine residues are all sensibly different, an ionization curve can be constructed for each, and the four peaks determined with good precision. Karplus *et al.* find, as did Masson and Wüthrich, that even in this alkaline range, some of the exchangeable peptide protons still remain unexchanged, and it seems that denaturation in these conditions is not wholly cooperative, and that even in so small a molecule independent cooperative units can remain tightly folded and resist the incursion of solvent.

Failure of cooperativity in denaturation, and the appearance of intermediates of long lifetime on the NMR time scale has now also been established for the archetypal case of ribonuclease. Thus Westmoreland and Matthews (*Proc. US Nat. Acad. Sci.*, **70**, 914; 1973), concentrating on the downfield histidine resonances—the separation of which because of the varied environments of these groups, is perhaps the most distinctive feature of the native spectrum—find that the acid denaturation profiles of the chemical shifts of these residues do not coincide. It is inferred that there are at least two separately unfolding regions in the structure, which is consistent with results of rapid kinetics of unfolding.

The reservation that secondary structure in general exists in acid-denatured proteins invests the guanidine hydrochloride system with particular interest, for here, as Tanford and his associates have shown, ribonuclease (and most other proteins) behave as random coils, possessed of no detectable intermolecular interactions. Nevertheless, here too the unfolding gives evidence of intermediate states when followed by NMR at high resolution. Benz and Roberts (*FEBS Lett.*, **29**, 263; 1973) have found that the downfield resonance from his-12 begins to decay at lower denaturant concentration than those of the other three histidines. When denaturation is complete only one C-2 histidine resonance, corresponding to all four groups in the coiled chain, is to be seen. His-12 occurs in the N-terminal "S-peptide" region, which is very possibly the most susceptible to disruption. This result is in a sense at odds with the kinetic and thermodynamic features of this system, which meet the criteria for a grossly two-state denaturation process, but as Benz and Roberts point out, the deviation from such behaviour in structural terms may be very slight.

PSYCHOLOGY

Depression and the Pill

from a Correspondent

AS many as one in fifteen British women on the contraceptive pill may find that it induces a state of depression which can be severe enough for the pill to be an unsuitable form of contraception for them. The reasons for this have been examined by a group at St Mary's Hospital Medical School, London, who have related the problem to a deficiency of vitamin B₆ (pyridoxine) in some women on the pill (P. W. Adams *et al.*, *Lancet*, **i**, 897; 1973). It seems that treatment with vitamin B₆ may sometimes be effective.

The biochemical basis of this work is that the administration of oestrogens interferes with the metabolism of the essential amino acid tryptophan, increasing the amount metabolized by the nicotinic acid pathway. The metabolism of tryptophan involves the coenzyme pyridoxal phosphate derived from vitamin B₆, and therefore a pill-induced increase in the rate of metabolism of tryptophan uses up pyridoxal phosphate.

But one of the essential amines in brain function, 5-hydroxytryptamine, is also produced by a pathway which requires pyridoxal phosphate, and in patients with depressive illnesses an examination of the fluid surrounding the brain and spinal cord shows this amine to be present in abnormally low quantities.

Attempts have been made in the past to treat premenstrual depression in women on the pill with pyridoxine, but the importance of the work at St Mary's is that for the first time a double-blind crossover trial with pyridoxine has been made.

One of the problems in selecting

patients for the trial was to ensure that the depression was a side-effect of the pill and did not have another cause. Most forms of depression give rise to symptoms of pessimism, tearfulness, anxiety and loss of sexual appetite, but also induce a disturbance of sleep pattern and appetite. These latter two symptoms are uncharacteristic of pill-induced depression, however, so women who had sleep and appetite disturbances or a history of mental disorder or obvious reasons such as marital disharmony that might lead to depression were excluded. The St Mary's team were then left with thirty-two women whose depression they felt to be caused by the contraceptive pill. Ten of these did not complete the trial, so the sample is small, but the results still seem significant.

The patients, divided into two groups, were treated with either 20 mg pyridoxine hydrochloride daily or a placebo and after two months the groups were interchanged. The women were on a variety of contraceptives though each contained 50 µg of oestrogen, and the length of time they had been on the pill made no difference to the results.

There was an alleviation of the depression in eleven women who at the start of the experiment were found to have an absolute lack of pyridoxine, but in the other eleven women whose biochemical assays were no different from other women on the pill the pyridoxine treatment showed no benefit over the placebo. So, as the St Mary's team point out, other causes must be sought for the depression in those women who did not respond to the pyridoxine.

Experiments along these lines may also conceivably throw some light on events in early pregnancy. Morning sickness has also been treated for some

Magnetic Effect of Cofactors

THE discovery that organic phosphates function as cofactors for haemoglobins, and are tightly bound to the deoxygenated state, has meant a re-evaluation of many physical properties, which were previously determined without regard to the presence and concentration of these ligands. The magnetic susceptibility of deoxyhaemoglobin, which is the subject of a re-examination by Alpert, Banerjee and Denis in next Wednesday's *Nature New Biology* (May 16), corresponds to ferrous haem iron atoms with an electronic spin of 2. This falls to zero in the liganded haemoglobin.

The classical value for the magnetic moment of deoxyhaemoglobin, obtained twenty-five years ago by Pauling and his colleagues, was about 5.4 Bohr magnetons. This turns out now to be the value for the complex with 2,3-diphosphoglycerate or inositol hexaphos-

phate. When the ligand is stripped off on a column, a drop to 4.9 Bohr magnetons is observed. This is exactly the theoretical value for independent spins. The higher magnetic susceptibility in the complex with cofactor must then be attributable to an added orbital contribution. The cofactor must therefore perturb the symmetry of the ligand field around the iron.

It is known from the X-ray work of Arnott that diphosphoglycerate binds in the central cavity of the protein at the junction of the two β chains, and in doing so induces displacements of up to 2 Å locally. Alpert *et al.* suggest that these can be sufficient to reduce the distortion of the ligand field around the iron from octahedral symmetry, which would have the effect of increasing the magnetic susceptibility. Both α and β chains probably contribute to this effect.

years with pyridoxine, so it may be that there is a relationship between morning sickness, increased oestrogen levels, and the depressions which can occur in early pregnancy.

HERPESVIRUSES

Genetics of HSV 1 and 2

from our Cell Biology Correspondent

THE genetic analysis of animal viruses, be they RNA viruses or DNA viruses, is still very much in its infancy but many animal virologists aspire after the achievements of bacteriophage geneticists and are seriously attempting to isolate suites of mutants that can be used in complementation and recombination tests. Animal virologists, of course, labour under severe disadvantages not shared by bacteriophage geneticists: animal cells grow slowly compared with bacteria, they are far more fastidious and to date nobody has detected suppressible mutants of animal viruses comparable to the amber and ochre mutants of phage. Temperature sensitive mutants of animal viruses, however, can be and have been isolated, and as Brown, Ritchie and Subak-Sharpe (*J. Gen. Virol.*, **18**, 329; 1973) take pains to stress, such mutants offer several distinct advantages which go some way towards compensating for their great disadvantage—their leakiness.

Subak-Sharpe and his colleagues have independently isolated nine 5-bromo-deoxyuridine-induced temperature sensitive mutants of herpes simplex type 1 virus, which because of its large genome (about 105×10^6 daltons of double stranded DNA), presents the animal virologist intent upon genetic analysis with a formidable task. These nine *ts* mutants apparently fall into eight complementation groups and, therefore, presumably define eight herpes simplex 1 cistrons. Moreover, because eight of the nine *ts* mutants selected at random fall into different complementation groups, Brown *et al.* estimate that the herpes simplex 1 genome probably contains more than thirty cistrons which specify functions that are indispensable for viral replication at 38° C.

Using these nine *ts* mutants and a plaque morphology marker in three factor recombination tests, Subak-Sharpe's group has also produced a tentative linkage map of the herpes simplex 1 genome. The map is linear, locates nine cistrons—the eight defined by complementation tests and the plaque morphology marker—and spans about twenty-five recombination units. In short, the group has made a useful start to the genetic analysis of this large and complex virus.

With another colleague, Timbury,

Subak-Sharpe has also begun to probe the range of genetic interactions between herpes virus type 1 and its relative type 2 (*J. Gen. Virol.*, **18**, 347; 1973). Making the most of their *ts* mutants of type 1 and type 2 viruses they have established not only that herpes simplex viruses types 1 and 2 can complement each other to a considerable extent, but also that the type 1 and type 2 genomes can recombine to produce stable recombinants. Furthermore, the progeny of cells infected with these two viruses include heterozygotes and in some cases viruses with a phenotype distinct from those of both parents. With the possibility that herpes simplex virus type 2 might be involved in the aetiology of human cervical cancer the Glasgow group should find it easy enough to justify extending this analysis.

Herpetologists will no doubt also have noticed Smith and de Harven's recent report (*J. Virol.*, **11**, 325; 1973) of what seems to be a neat method for concentrating stocks of herpesviruses, many of which do not yield high titres when they replicate in cultured cells. Apparently negative pressure ultrafiltration through dialysis tubing does the trick, by converting low titre stocks into high titre concentrates of intact unaggregated virus particles without loss of infectivity.

CELL FREE SYSTEMS

Translation *in vitro*

from our Cell Biology Correspondent

THE person who first manages to isolate strains of cultivated animal cell which harbour suppressor mutations and as a result contain suppressor transfer RNAs

capable of translating amber or ochre codons in messenger RNAs will have achieved a triumph. The opportunity to isolate and exploit suppressible nonsense mutants of animal viruses is something animal virologists long after, and until suppressor mutant cells are isolated virologists must content themselves by working with temperature sensitive mutants that are a deal less satisfactory. The isolation of suppressor strains of animal cells is, it scarce needs saying, a formidable task that promises much hard and tedious work with no guarantee of ultimate success and, not surprisingly, nobody has embarked on it seriously. But thanks to the efforts of Schreier, Staehelin, Gesteland and Spahr (*J. Mol. Biol.*, **75**, 575; 1973) there is now at least an *in vitro* assay system that can be used to screen for the suppression of nonsense mutants in eukaryotic cell extracts.

Schreier and Staehelin (*J. Mol. Biol.*, **73**, 329; 1973) have recently devised a highly efficient mammalian cell free system in which, for example, during 40 min at 30° C, each ribosome will translate a globin messenger RNA molecule three or four times. While Gesteland and Spahr during the past several years have made themselves masters of *Escherichia coli* cell free systems which translate efficiently not only the genomic RNAs of RNA bacteriophages such as R17 and Q β , but also the messenger RNAs of T-even phages. These workers in collaboration have established that the mammalian cell-free systems of Schreier and Staehelin can, with appropriate modifications, be induced to translate faithfully the coat protein cistron and the RNA replicase cistron

Messenger Translation in *Xenopus* Oocytes

A CAUTIONARY tale is told in a short contribution to next Wednesday's *Nature New Biology* (May 16) presented by Lane, Gregory, Iyatsumi and Scherrer. In recent years, evidence for the existence of messenger RNA sequences in high molecular weight RNA (HMW RNA) has been reported, based on experiments in which synthesis of globin or immunoglobulin occurred following injection into *Xenopus* oocytes of HMW RNA extracted from the appropriate tissues (erythroblasts or myeloma cells). This made a neat and happy story, but now Lane *et al.* plead caution. The trouble seems to lie in the extreme sensitivity of the oocyte system as a test for mRNA translation. Thus, if as little as 1 per cent of the HMW RNA injected into oocytes is represented by contaminating aggregated cytoplasmic messenger, then synthesis of the appropriate protein would be detected. Even excluding cytoplasmic contamination, it is possible that nuclei,

from which the HMW RNA is extracted, may contain functional mRNA.

The control experiment carried out by Williamson *et al.* (*Nature New Biology*, **241**, 66; 1973) to eliminate the possibility of 9S globin mRNA contamination of their HMW mouse erythroblast RNA is inconclusive. Williamson *et al.* demonstrated that no globin was synthesized when brain HMW RNA (extracted from a mixture of brain tissue and reticulocyte lysate) was injected into *Xenopus* oocytes. Lane *et al.* point out, however, that 9S mRNA in the lysate may have been degraded by brain enzymes. They suggest a control experiment to solve the problem. If HMW RNA were extracted from a mixture of duck erythroblast nuclei and rabbit reticulocyte polysomes then, if messenger aggregation does not occur, the HMW RNA should code for duck but not rabbit globin, whereas the 9S fraction from the same preparation should cause the synthesis of both globins.

of Q β or R17 phage RNA. What is more, they have found that when the mammalian cell free system is programmed with RNA from the R17 amber mutant *amb2*, which carries an amber mutation at position 6 in the coat protein cistron, no coat protein is made. In other words, this amber mutation terminates faithful translation of the coat protein cistron in the mammalian cell free system just as it does in *E. coli* cell free systems. Obviously *amb2* R17 RNA can now be used to test cell free systems prepared from various animal cells by Schreier and Staehelin's methods to see if any contain an amber suppressor tRNA.

Virologists and others would also clearly like to have a cell free system which faithfully translates single stranded DNA from virus particles and cells because it is far easier to isolate and purify viral DNA than viral RNA. Since the mid-nineteen-sixties various groups have tackled this problem only to meet with disappointment. Bretscher (1969), for example, found that an *E. coli* cell free system programmed with phage fd DNA made a set of fMet dipeptides many of which were not related to known N termini of the phage proteins. Ihler and Nakada (1970, 1971), on the other hand, obtained more encouraging results: in their hands ribosomes appeared to bind to authentic initiation sites in T7 and T4 single stranded DNA. But according to Cardit, Goldberg and Steitz (*J. Mol. Biol.*, **75**, 449; 1973) ribosomes in conditions which allow optimal RNA-directed chain initiation in an *E. coli* cell free system, bind to single stranded DNAs from eight unrelated viruses or cells to yield predominantly three dipeptides fMet-

Thr, fMet-Val and fMet ^{Leu}_{Ileu}. As

they say, "although dipeptide synthesis primed by natural DNA templates may yet prove useful in deciphering the molecular basis of protein synthesis, it is probably of little value as a tool for decoding genetic information *in vitro*".

INERT GASES

Elastic Constants

from a Correspondent

THE adiabatic elastic constants of single crystals of neon at 24.3 K and of argon at 82.0 K have been determined by Stoicheff and his colleagues at the University of Toronto, using the Brillouin light scattering technique (*Phys. Rev. Lett.*, **29**, 1454; 1972). The results represent what is probably the most accurate set of elastic constant data currently available for the solidified inert gases, and they are important be-

cause they provide a sensitive test of recent theories of lattice dynamics. The measurements are particularly useful because they are made near the triple points, a region about which there are unresolved questions concerning the influence of anharmonicity and many-body forces.

The inert gases crystallize in face-centred cubic lattices in which the atoms are bound by the cohesive van der Waals interatomic forces. These forces are central and may be represented to a good approximation by simple radial potentials. Thus the solidified inert gases correspond to elementary models for the solid state and are therefore ideal for developing the lattice dynamics of simple solids. These theories have to be tested against experimental data for the bulk properties of the crystals. Most experiments so far on the solidified inert gases have, however, been carried out on polycrystalline samples and only recently has it been possible to prepare large single crystals. Some data for single crystals are now available for the lattice parameter, isothermal compressibility, thermal expansion and other thermodynamic properties; but to investigate properly the dynamical properties of the lattices, and hence the interatomic forces, accurate measurements of the elastic constants are also required.

Because of its small atomic mass and the consequently large amplitude of its lattice vibrations, neon provides a very stringent test of theory. Before the work of Stoicheff *et al.*, however, only one other determination of its elastic constant had been reported, based on neutron scattering on two crystals at 4.7 K. Argon, on the other hand, is the most thoroughly investigated of the solidified inert gases, both theoretically and experimentally. Even in the case of argon, however, there is considerable disagreement between the various experimental values for the elastic constants and a similar disagreement between experimental and theoretical data.

In spite of the improved techniques now available for preparing single crystals, accurate measurement of the elastic constants is difficult. Determinations of acoustic velocities in specific crystal directions have been made using neutron scattering and ultrasonic techniques. But so far these methods have yielded results which not only are mutually inconsistent but also imply a much lower elastic anisotropy for the crystals than can be accounted for theoretically. The principal difficulty with both neutron scattering and ultrasonic techniques seems to be that large single crystals (~ 1 cm³) are required; these are hard to produce and maintain under the appropriate experimental conditions.

Recent advances in high resolution

Brillouin spectroscopy now allow values of adiabatic elastic constants for crystals to be determined with high precision by this technique. The application of the method to the solidified inert gases has two important advantages over the alternative techniques: first, because it only requires small samples (~ 1 mm³); and, second, because light scattering studies do not require a transducer or other device to be physically bonded to a crystal sample.

Stoicheff *et al.* have grown crystalline samples of argon and neon in the tail sections of suitable dewars and have checked orientation and quality of the crystals by means of Laue X-ray transmission photographs. Brillouin spectra were recorded for the crystals at several different orientations, using a single frequency argon ion laser and a piezoelectric scanning interferometer. The observed Brillouin spectra were found to contain the longitudinal component and one "slow" transverse component. The reason for the non-appearance of the expected "fast" component is that it is generally of extremely low intensity in the solidified inert gases, except for a limited range of orientations not included in the present arrangement. Each of the frequency shifts, ν_i , was subsequently analysed using the usual Brillouin equation

$$\Delta\nu_i = \pm 2 \nu_i (V_i/c) n \sin(\theta/2)$$

to determine the velocities V_i of the thermal waves.

Three independent adiabatic elastic constants C_{11} , C_{12} and C_{44} describe the elastic properties of a cubic crystal. Values of these constants accurate to about $\pm 4\%$ were deduced from the velocities, together with values for the adiabatic bulk modulus and the Grüneisen parameter.

The results for neon at 24.3 K are all 25–40% lower than the only other set of data available for neon, at 4.7 K. This suggests that anharmonicity is very important in neon. The results for argon differ quite considerably from earlier investigations reported, particularly from measurements based on ultrasonic techniques. The reason for this is presumed to be because earlier measurements have been carried out chiefly on polycrystalline specimens. The great value of the present method lies in the knowledge that the samples were single crystals of well defined quality and orientation.

It is somewhat surprising to find that the experimental values of C_{11} and C_{12} still differ from the calculated values by about 1.5 times the estimated uncertainties. This is, however, much better agreement than has generally been found in the past. At this stage, it would be very useful to have further measurements on argon and neon, particularly at different temperatures.

GRAVITATIONAL RADIATION

Waving Goodbye to Weber's Waves

by our Cosmology Correspondent

WEBER'S gravitational wave detectors may well be detecting something—but it is almost certain that they are not detecting pulses of gravitational radiation. This was the clear consensus that emerged from the Oxford-Cambridge-London Relativity Seminar, held at All Souls College, Oxford, on April 27. Theoretical discussions formed an important part of the seminar, particularly those concerned with the possibility of detecting black holes and the implications of the discovery of Cyg X-1, a binary system which contains the best candidate yet for such a completely collapsed object. But the proceedings were dominated by the experimentalists and their reports of the present status of the search for gravitational radiation—a remarkable turnaround from the situation a couple of years ago, when it would have been laughable to suggest that such a relativity seminar could be so concerned with experiments.

Dr R. W. P. Drever (University of Glasgow) summarized the present situation in the experimental field. All the present crop of experiments were, of course, inspired by Weber's claims that pulses of gravitational radiation were being detected by his equipment. Others have found it very difficult to duplicate Weber's results, and questions regarding the sensitivity of his equipment, and the possibility of unconscious human bias, have inevitably arisen. A team from the Soviet Union were the first to follow Weber, using a detection system which had the advantage that it could be accurately calibrated.

Basically, all the gravitational wave detectors consist of massive aluminium cylinders, cooled to reduce thermal effects and suspended in a vibration-free environment. Weber's equipment has never been calibrated directly; he estimates its sensitivity as better than 1 *kT*, but others have estimated the response of this apparatus as only sensitive to 10 *kT* pulses. The Soviet instrument is calibrated as sensitive at the 9 *kT* level, so some doubt remains as to whether that instrument's negative result should be directly compared with Weber's positive results.

What of more sensitive detectors? Basically, there are two approaches now being employed: bigger detectors and/or more sophisticated signal processing. Neither approach has met with success in the form of a definite detection of even one pulse of gravitational radiation. At the University of Bristol, what is essentially a second generation experiment, in terms of sensitivity, is still being constructed. There, the attitude is that enough people around the world are

already producing negative results at the few *kT* level, and that it is worth waiting a little longer to produce a machine more sensitive by five orders of magnitude. In Glasgow, a detector rather smaller than Weber's but with about the same sensitivity is one of the many producing negative results, and, perhaps most interesting of all at present, a joint Munich/Frascati system, designed to be as similar to Weber's as possible, is also failing to produce any positive results.

The conclusion, from these and other experiments, seems inescapable. Weber has not found pulses of gravitational waves. Dr Drever speculated that perhaps Weber's pulses are associated with disturbances in the Earth's magnetic field, because his apparatus is unique in containing large amounts of ferrous material, although the detector cylinder itself is non-magnetic. It is also still just possible that Weber has been fortunate in building a detector which operates at a favoured frequency for gravitational radiation, but this faint prospect did not stir any enthusiasm in Oxford.

The theoreticians were far from displeased to learn of these negative results, because according to general relativity sensible disturbances, such as supernovae

explosions, should produce gravitational radiation detectable on Earth only with equipment much more sensitive than Weber's. Professor M. Rees (University of Sussex) estimated that an improvement by a factor of 10^8 is needed before positive results can be expected, because this would extend the range of the detector to the Virgo cluster and offer hope of detecting one supernova a year. Professor D. Lynden-Bell (University of Cambridge) turned the question around to ask at what level of sensitivity relativists would be distressed to learn of the continued failure of the experiments to find gravitational waves. It seems that the pundits will only begin to worry when negative results are obtained by experiments 10^{12} times more sensitive than those of today.

That might have seemed to have brought the young science of gravitational wave experimentation to an untimely demise; but Dr Drever promptly returned to the fray to suggest that at the present rate of improvement in techniques the first quantum of improvement, by Professor Rees's factor of 10^6 , could be achieved within a few years. As well as the ultimate refinement of the Weber approach, a 5 to 6 ton cylinder levitated by superconducting magnetic coils and maintained at 1/30 K, with tiny oscillations detected by Josephson junction effects (with instru-

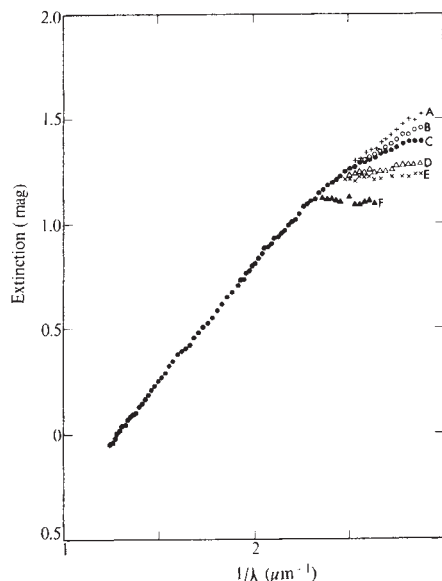
Reddening of Stars near the Galactic Centre

IN next Monday's *Nature Physical Science* (May 14) Whittet, van Breda and Nandy describe interstellar extinction curves which they have derived from photoelectric scanner observations of twenty-two early type stars in a region of the southern Milky Way which includes the galactic centre. There are considerable variations in the data, which are not correlated with position of the stars in the plane of the Galaxy, suggesting that the extinction law in these regions is strongly influenced by the physical conditions which prevail in local dust clouds.

The scanner observations were made with the 36-inch telescope at Cerro Tololo during May 1972. They covered 3480 to 5240 Å with a bandwidth of 40 Å in the second order, and 5160 to 8040 Å with a bandwidth of 80 Å in the first order. The area surveyed was from $l^I=260^\circ$ to $l^I=30^\circ$, passing through the galactic centre.

The twenty-two reddened stars examined seem to fall into six chief groups, as far as the extinction curves are concerned (see figure). But the stars in each group are not spatially related—for example, stars which contribute to extinction curve D include one at $l^I=343^\circ$, one at 15° , one at 263° and three others. Even this

distinction is somewhat artificial, however, and Whittet *et al.* point out that there seems to be a continuous family of curves, at least between curves A and D in this figure. Of the nine stars observed in the galactic centre region "six show a reddening law close to curve C; the other three follow curves A, B, and D".



Characteristic extinction curves for reddened stars observed at Cerro Tololo.

ments being developed jointly by a Stanford/Louisiana State University/Rome group), there is also the prospect of experiments in space with masses a long distance apart, distances being monitored by laser interferometry. Weber may not have detected gravity pulses, but by inspiring the work of the past two years around the world he has ensured that there is now a real prospect of making the final push to produce detectors sensitive at the levels which relativity theory suggests will be suitable for the detection of such radiation. Just as the steady state theory provided the incentive for work which ultimately led to convincing evidence in favour of big-bang models, so Weber's incorrect interpretation of the events detected by his apparatus may prove the first step in the detection of pulses in line with relativity theory.

TROPICAL ECOLOGY

Fire and Savanna

from a Correspondent

IN 1952 P. W. Richards (*The Tropical Rain Forest*, Cambridge University Press) expressed the general view that the savanna grasslands of Africa, and their recent rapid spread south of the Sahara at the expense of closed forest, are wholly ascribable to the increased incidence of man-induced fire rather than to climatic conditions and a recent increase in aridity. These African savannas have been described as the largest area of mismanaged land in the world, yet fire can be exploited judiciously as a management tool. On April 9 the Tropical Group of the British Ecological Society met to discuss recent research on the effects of burning on savanna vegetation and animal populations.

Dr R. Rose-Innes (Land Resources Division, Overseas Development Administration) described how firing early in the dry season induces a mild burn favoured by silviculturists; the intense late burns encourage grassland at the expense of forest, and burns after the first rain can destroy the woody vegetation as it begins to shoot. Dr R. M. Lawton (Land Resources Division, ODA) elaborated the use of fire in forest management; Dr R. H. Kemp (Commonwealth Forestry Institute, Oxford) pointed out, however, that savanna regions could provide 10–20 times their present yield per hectare of hardwood timber if planted with tropical pines. The use of photogrammetry for interpreting patterns of savanna burning in relation to climate, soils and agricultural practice was summarized by Dr R. P. Moss (University of Birmingham), and Dr A. Blair Rains (Land Resources Division, ODA) described its use for estimating grazing

resources in the semi-arid regions of Southern Africa where grass is too sparse to support a fire detrimental to woody growth.

Professor M. R. Delany (University of Southampton) and Dr B. R. Neal (University of Saskatchewan) studied the effect of fire on small rodent populations in Uganda's Queen Elizabeth Park. Twelve species were found, if considered by three size categories, to occupy specific niches defined by micro-habitat, food resources and feeding behaviour; responses to fire differ, but there is an initial increase in numbers, after which a drastic decline and eventual recovery follows. Small mammals favour the cover of tall grasslands, and their numbers and diversity are inversely proportional to those of large mammals which favour the more nutritious short grasslands.

Contributions by Professor D. H. N.

Spence (University of St Andrews) and Dr G. Harrington (CSIRO, Canberra) on burning and grazing in relation to deterioration of habitat diversity in two Ugandan national parks emphasized the impression gained at the meeting as a whole: fire, the old scourge of African savanna, is being overtaken by the new spectre of overgrazing, especially by elephants. In parts of East Africa fire exclusion leads to no appreciable amelioration of deforestation unless grazing is also excluded; the old traditional use of fire-arms, and the prevalence of tsetse, in West Africa has so far prevented overgrazing, but with the elimination of tsetse it can be anticipated. Dr Kemp's dream of productive softwood forestry in African savanna therefore remains a long way off, while in the Brazilian savannas the new highway is leading to a rate of forest devastation never before witnessed.

More Very Old Rocks from Greenland

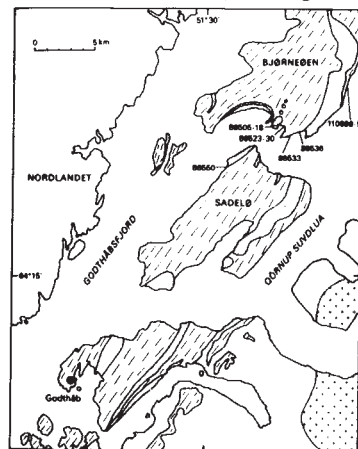
THE oldest known rocks in the world are quartzofeldspathic gneisses (the Amitsoq gneisses) from the Godthaab area of West Greenland which were dated in the range 3,700–3,750 million years by members of the group at the University of Oxford in collaboration with the Geophysical Survey of Greenland (Black *et al.*, *Earth Planet Sci. Lett.*, **12**, 245; 1971 and Moorbath *et al.*, *Nature Physical Science*, **240**, 78; 1972). In next Monday's *Nature Physical Science* (May 14), members of the same team (Pankhurst *et al.*) report further rubidium–strontium ages from this part of Greenland, although the new ages represent a more recent event in the evolution of the Godthaab area.

The rocks in question are the Nûk gneisses which lie in close association with Amitsoq gneisses but which occur as dykes and subconcordant sills cutting the older rocks and as large subconcordant complexes. Most of the samples now dated by Pankhurst and his colleagues are from a large polyphase complex of quartzofeldspathic rocks in which the earlier phases are dioritic and the later ones granitic. The interpretation given for the Nûk gneisses is that they are derived from a syntectonic suite of calc-alkaline igneous rocks which were intruded into the Amitsoq gneisses and the Malene supracrustals (metavolcanics and metasediments) which postdate them. Conversion to gneisses then took place by metamorphism and deformation both during and subsequent to, the intrusion of the parent rocks.

The Nûk gneisses dated are from the north of Godthaab where they are metamorphosed to the same degree as the associated Amitsoq gneisses and Malene supracrustals. The metamorphic grade

is, however, significantly lower than that to the south of Godthaab where the 3,700–3,750 million year old Amitsoq gneisses were obtained. For this reason the rubidium–strontium isochron age for the Nûk gneisses is thought to represent the time of crystallization of the igneous parent material rather than the time of later isotopic rehomogenization associated with metamorphism.

The age obtained by Pankhurst *et al.* is $3,040 \pm 50$ million years, which gives another fixed point on the time scale for the crustal evolution of West Greenland. Thus the plutonic development giving rise to the parent rocks of the Amitsoq gneisses, the conversion of the parent rocks to gneisses, the intrusion of a basic dyke swarm (the Ameralik dykes) into the gneisses, the production of the Malene supracrustals, and the intercalation of the Amitsoq gneisses and the Malene supracrustals, must all have taken place in the interval from about 3,700 to 3,000 million years ago.



The Godthaab region of West Greenland. The Nûk gneisses are represented by diagonal dashes, the Amitsoq ones by the stippled pattern.

"Backcasting" with the World Dynamics Models

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Two members of the Science Policy Research Unit at the University of Sussex discuss whether or not a Systems Dynamics model can meaningfully be run backwards in time.

THERE seems to be some misconception about both the feasibility and the utility of running a System Dynamics model backwards in time (see, for example, Dennis Meadows's response to the Science Policy Research Unit's critique of his World Dynamics model in *Futures*¹). That issue forms the principal substantive technical criticism of our work made by Meadows. By clarifying this issue once and for all, we would hope that arguments about methodological technicalities, although important, will not be allowed to obfuscate the very important and more fundamental criticisms of his work which are made in the *Futures* articles.

Equating

With regard to the issue of feasibility, it is instructive to consider the kind of equations generally used in such models, and the circumstances in which it may not be appropriate to "backcast" using such a model. System Dynamics models comprise a set of non-probabilistic, non-linear first order difference equations. Using the Dynamo programming language they are solved numerically over discrete time intervals. What one is attempting to do by this is to approximate to the continuity of the set of assumed mechanisms which make up the model. The equations may include feedback processes (that is, the rate of change of a variable depends at least in part on its own value) and delays (that is, a change in a variable does not instantaneously affect any others). The equations used to introduce delays in the System Dynamics method also take the form of first order difference equations (they do not represent true delays). As the original system is assumed to be continuous there is no mathematical reason why the properly posed model should not run as well in the backwards direction as it does in the forwards direction. Provided that the parameter values are correct, any inability to reproduce transients when calculating through the same time period from opposite directions must therefore arise as a result of the approximations made during the computer calculation of a System Dynamics model using the Dynamo language.

In the first place it should be pointed out that the order in which computer calculations are carried out is not perfectly reversed when the sign of the time step used in the calculation is switched. In the forward calculation using, for example, a step length between successive calculations of one year, parameter

values for the year (say) 1930 are determined from those at 1929, whereas in the backwards direction they come from those at 1931; because in most dynamic systems rates of change are not constant, this leads to asymmetry when calculating in opposite directions. Provided the step between successive calculations can be made small enough, any errors of approximation (as opposed to numerical errors) arising from this ought always to be insignificantly small. If, however, one reduces errors at each iteration by reducing the time increment to a very small value, the total number of iterations is increased and sooner or later numerical errors will still accumulate adversely. A related problem arises when numbers generated by the equations of the computer model are of the same order of magnitude as the rounding errors involved in the numerical computation (the Dynamo language reads numbers to five significant figures only, although it calculates to more). This can occur, for example, when the difference between two very nearly equal numbers is taken. In such cases the numerical solution to the equations of the model will not agree with their analytical solution. The level of agreement needed depends on the problem, but certainly the numerical accuracy should be such as not to mislead. It is extremely important, therefore, in the development of a dynamic model to ensure that at no point in the calculation are solutions excessively interfered with by either numerical or approximation errors. Further, unless this is assured, solutions calculated through a particular time period from opposite directions will not be the same. If these aspects are properly attended to there should be no reason why trends evaluated in the forwards direction should be any closer to the analytically derived solution than trends calculated in the reverse direction.

There is, however, a further danger if the solutions of a dynamic model are matched to data from a period in which only monotonic trends have been observed; potential errors are automatically built into the model, for parameters must be "adjusted" to account for the systematic biases of approximations in the calculation. Consequently when the model is reversed or used to forecast into a period for which the "adjustment" is no longer valid, approximation errors may be more prominent. As an aside it should be pointed out that in most System Dynamics models where transients are not monotonic, individual relationships are actually taken as time reversible even when calculating in the forward direction.

There are, of course, circumstances in which a dynamic model should not be expected to run backwards. For instance, it is well known that certain difference equations have different numerical stabilities depending on the direction of calculation (for example, Bessel's difference equation for some parameter values). Another example would be a model in which any process within the model has decayed as a result of forward running to the extent that errors of approximation are already affecting parameter values. In some models, but not in the System Dynamics models known to us, transients are continually decaying and being replaced by others (for example,

models of turbulent flow which are usually representations of a steady state) and here it is not appropriate to reverse the time direction of the numerical calculation. For this reason, it is certainly not correct, as Anthony Tucker has done², to draw analogies between System Dynamics models and models of aircraft flight.

With regard to the utility of running a System Dynamics model backwards, the following may be said. As a model is a mimic of reality, it should reflect mechanisms and trends for the period of interest. If the same mechanisms were important in the past (before that period of interest), it is only reasonable to expect that a model should "backcast" or extrapolate into the past. In some sciences, for example, astronomy, retrodiction is as important as prediction. To argue that one may not backcast using a model is to say that mechanisms were in fact different in the past. This is an essential part of the scientific paradigm and applies equally in both the physical and the social sciences (see ref. 3). To argue that one should not attempt to backcast using a System Dynamics model, while at the same time admitting that the same mechanisms operated in the past, is to say that such models are beyond the reach of normal scientific criticism. The same can be said of a model for which the individual relationships are taken as reversible in the forwards direction. If one accepts that System Dynamics models are amenable to normal scientific criticism then "backcasting" clearly has a certain utility. In a non-linear dynamic model containing many parameters and many feedback loops, it is not practical to make all possible tests on the reasonableness of the model. "Backcasting" can be expected to drive some variable to unrealistic values and hence show up inadvertently incorrect assumptions underlying a model. Besides this, successively running a dynamic model forwards and backwards about a given point in time will point to any inconsistent assumptions and numerical instabilities within the model. This technique has been successfully employed in the construction of the ILO Bachue model⁴.

World Dynamics Models

In the light of the above discussion, it is now pertinent to turn to the World Dynamics models of Jay Forrester and Dennis Meadows and the ongoing debate between Meadows and the Science Policy Research Unit (SPRU). In our analysis at SPRU of Forrester's and Meadows's System Dynamics world models, we examined the performance of the models when run backwards in time from their starting date (1900). The reasons for doing this were as follows: the (World 2) model exhibits unrealistic population growth in the immediate post-1900 period (even though its parameters have been adjusted to match 1900 to 1970 world trends). This was not apparent from the publication *World Dynamics*⁵ presumably because of the coarse scale used to illustrate Forrester's runs of the model. The population figures calculated at 1904 are actually lower than those calculated at 1900. At 1900 the assumed birth and death rates are reversed in magnitude. Their values are 48 per thousand and 55 per thousand respectively which compares rather poorly with the accepted 1900 figures of about 32 and 20 for the developed regions of the world and 40 and 35 for the under-developed regions⁶. It is clearly instructive to extrapolate these apparent errors to an earlier period to see where the trends generated by the model lead from. Further, as already pointed out, running a model backwards can give some indication of its reliability in the forward direction.

As explained in *Futures* and in ref. 7 running World 2 backwards to before 1880 proved impossible, because of instabilities arising principally from poorly estimated parameters used in the calculation of birth and death rates. For obvious reasons only the essence of our argument concerning this effect was presented in *Futures*, although it was fully communicated to

Meadows at a date well before publication. In view of Meadows's preoccupation with this technical detail, however, we are obliged to discuss the matter further. But first, there are a number of "red herrings" to be dealt with. Meadows says, for example, that "reversing the sign of the solution time increment to make a feedback model 'run backwards' must radically alter the entire dynamic character of the model. With a negative time increment, negative loops are converted into positive ones (for example population will be augmented by deaths and depleted by births)". We fail to see the relevance of this obvious truism. He says "delayed relationships will also be asymmetric in time under some circumstances" and that this is another factor which makes backcasting with the models "completely meaningless". The fact is, however, that there are no explicit delays in Forrester's World 2 model. Those in World 3 may be reversed in the same manner as other equations in the model simply by making the sign of the time increment negative. Furthermore, Meadows says, "stochastic influences on the behaviour of a model in the forward direction will not, in general, be recaptured in the reverse mode". But the fact is that neither Meadows's nor Forrester's world models are probabilistic. None of the equations account for random events. Similarly Meadows's remarks about our forward projections with the World 2 model from 1850 do not apply to what we actually did.

Meadows says (in his response in *Futures*), "disequilibria introduced through the assignment of initial values may insert transients in the model's behaviour which cannot be reproduced in the reverse direction". The assignment of inconsistent initial values can cause confusion in dynamic models, especially those pertaining to the social sciences where in general theories and data sources are relatively weak and often inconsistent. Inconsistency between the initial values given to the parameters of a dynamic model and the set of equations which link them may arise. In successive iterations of the dynamic calculation the parameters will adjust to the form of those equations and the model will "settle down". Where the equations and parameters are inconsistent in the same sense that a set of simultaneous equations are inconsistent "settling down" can be rapidly achieved by beginning the iteration with a very fine time increment. The effects of wrong structural assumptions within a model, on the other hand, are not removed by this technique (see the discussion of the World 2 population minimum in *Futures*).

Sussex Experiment

In the Sussex experiment the World 2 model is run backwards from 1900 and forwards again through 1900 giving the same qualitative results at each pass. The calculation of "pollution" exhibits numerical instability when running in the backwards direction and Meadows shows that in attempting to run the World 2 model backwards from 1940, errors of the order of one part in 10^5 in the initial values (that is, of similar order of magnitude to the Dynamo approximation errors) can cause the model to be unstable. He implies that this same instability invalidates our comments on "backcasting" from 1900. This is simply not the case—although the instability is present it drives the net pollution to negative values which are then taken as zero, thus effectively decoupling pollution from the rest of the system. The insertion of more realistic values of pollution had only very slight quantitative effect on the behaviour of the other parameters. Meadows has shown that small biases can be introduced into the pollution sub-system when backcasting from 1940 which can result in pollution taking large positive values and that this drives other parameters to abnormal values. Unfortunately we are unable to reproduce the effect shown in Meadows's response without the introduction of such biases. He might equally well have shown runs with an opposite bias to the pollution system. In this case the model would retrace its population trajectory back-

wards through 1900 and the retrodicted population "explosion" in 1880.

Finally, on this point, Meadows says that if the state variables are entered with "absolutely no error" (as with the technique adopted originally with Bachue) then World 2 can backcast its own behaviour over limited periods even from 1940. (Running the model backwards from 1900 poses no problem in this respect—the initial values are identically those used for the forwards run.) Whereas the instability in the backward run from 1900 is shown to derive largely from wrong assumptions in the model, that at 1940 comes from numerical errors in the computation. The variables which are most affected are POLG (pollution generation) and POLA (pollution absorption). These are calculated quite independently. In 1940 these take the values 1.737 and 1.701 units respectively, so changing the numerical values of any one of the several parameters used to calculate them by about 2% can turn a net pollution increase into a decrease when calculating in the forward direction. As no information of such accuracy is available for "pollution" (see, for example, Marstrand and Sinclair in chapter 7 of ref. 7) the population collapse resulting from the accumulation of pollution which is "predicted" by the model must be viewed with some reservation.

In *The Limits to Growth* Meadows⁸ says "... every assumption we make is written in precise form so that it is open to in-

spection and criticism by all" and "implications [of assumptions] can be traced without error by a computer no matter how complicated they become". To the first statement we would comment that the equations of Meadows's model (World 3) were still not generally available one year after his book was published. To the second we would argue that it is the responsibility of the modeller of a complex system not only to show awareness of the hazards of computer modelling but also to demonstrate that they do not invalidate the conclusions which are drawn.

By answering Meadows's criticism and by drawing attention to the issues involved in a particular aspect of modelling, "back-casting", we are pointing to some of the difficulties and dangers inherent in building complex computer models. Finally, we would reiterate our hope that the arguments can now turn back to the more fundamental issues.

¹ *Futures*, 5, No. 1 (1973).

² *The Guardian*, March 7 (1973).

³ Brown, Robert, *Explanation of the Social Sciences* (Routledge, London, 1963).

⁴ *Nature*, 241, 160 (1973).

⁵ Forrester, J. W., *World Dynamics* (Wright Allen Press, Cambridge, Mass., 1971).

⁶ *A Concise Summary of the World Population Situation in 1970* (United Nations, 1971).

⁷ *Thinking about the Future* (Sussex University Press, 1973).

⁸ Meadows, D. L., et al., *The Limits to Growth*, 22 (Universe Books, New York, 1972).

Mathematical Literacy for the Non-mathematician

GEORGE STEINER

Churchill College, Cambridge

This article is based on an address by Dr Steiner at the annual award ceremony of the Guinness Prizes in Mathematics and the Sciences in July 1972.

THERE is a well known passage in a letter of Keats in which the poet says how ardently he would wish to know precisely how Shakespeare sat or moved about the room when he was composing "To be or not to be" or another of his immortal soliloquies. I have often wondered how R. Clausius sat when, in a paper on thermodynamics of 1852, he gave the first definition of entropy. The concept had been implicit in the study of caloric theory since Sadi Carnot's *Reflections* of 1824. The mathematical bases had been worked out in Clapeyron's *Memoir on the Motive Power of Heat* ten years later. Kelvin had grasped and publicized the importance of Carnot's analysis. But it was Clausius who, with a characteristic German bent for the total metaphysical picture, drew the epochal conclusion. The Universe is running down; the increase of entropy is ineluctable. What lies ahead is, in Clausius's later term, a "heat death" or the inertness and randomization of total thermal equilibrium. The notion that the Universe will end either in conflagration or lifeless cold, is as old as mythology. Structurally it probably derives from the fact that our intellect is scarcely able to conceive of a beginning without an end.

But until Clausius, the force of the idea was symbolic; it belonged to the domain of apocalyptic or messianic clichés. In Clausius's papers on thermodynamic exchanges and the theory of gases, the image of an end to our universe takes on the quiet certitude of an algebraic function. It is seen not as a religious mystery or philosophical metaphor: but as a mathematically verifiable truth (and the necessary algebra is, in fact, of an elementary elegance, deriving ultimately from $e = T \log v$, where e is the quantity of heat needed to maintain the temperature of a gas during dilatation, T the temperature in degrees and v the volume in litres). In short, Clausius wrote down the proposition, and the proof of the proposition, that there will be an analytically demonstrable, analytically foreseeable end to time and to the cosmos. Did he put down his pen for a moment or go to the window and look out?

It would be foolish to set the one moment above the other. Clausius's sentence generates the magic of exact truth. The precisions and truths in a Shakespeare soliloquy are of another order. Keats himself felt that the two classes could be compared and, in a somewhat gnomic passage, makes of "truth" an eagle higher than "poetry". But it is more useful to see at how many points the two modes of achievement correspond. Both represent the ultimate pressure of the human mind on reality, the capacity, indeed the compulsion, of consciousness to generalize beyond the discrete fact, to construct imaginatively and to impose on the world a living shapeliness. The poet and the mathematician are the great refusers of disorder. They use language—and mathematics is, above all, a language—to form constructs of meaning, of organic relation, of rational prediction without which our

sensibilities would operate at those monotonous levels of instinctual immediacy, of constantly awed acceptance which seem to delimit the mental existence of animals. Poetry, philosophy, mathematics are the counter-statement of man to the brute determinism of the organic condition. We owe to them the best of our dreams. In poetry and mathematics, moreover, the key to major forms is one of disinterest. G. H. Hardy's axiom of "uselessness" is an extreme statement. The "uses" of poetry and of even the purest mathematics are manifold, but of a special kind. They provide the psyche with free spaces, with an indispensable sensation of freedom in the face of biological or social constraints. A play by Shakespeare, a paper by Gauss, are an essential reminder of the freedom of thought from the constraints of reality, from the demands of mere survival. This last point is perhaps the deep link. The great poet gambles on transcendence, on the uncanny power of words to live far longer than those who have first spoken them. Clausius affirms that the human intellect can, via the unsparing dignity of consequent reason, foretell an end to the universe which it inhabits. No less than the artist, the mathematician can answer with Jacobi, that he practices his craft "for the honour of the human spirit", that he comes as near as we can to showing the triviality of death. Hence a quality of delight common, vital to both. "If I was smitten by algebra", wrote Edgar Quinet, "I was dazzled by the application of algebra to geometry. . . . The idea, the possibility of expressing a line, a curve, in algebraic terms, by an equation, seemed to me as beautiful as the *Iliad*. When I saw this equation function and solve itself, so to speak, in my hands, and burst into an infinity of truths, all equally indisputable, equally eternal, equally resplendent, I believed I had in my possession the talisman which would open the door of every mystery." The "zero at the bone" should be the same when first we hear a great line of verse—St John of the Cross's *Oh noche amable más que el alborada*—and when we first grasp Cantor's proof that there is no greatest cardinal number. In either moment, the mind has stretched and altered the measure of man.

It is when the two modes interact that culture acquires a special lustre and coherence. Precisely this interaction characterizes the two seminal moments in western intellectual history, the flowering of ancient Greece and the period from Galileo and Descartes to Newton. It is impossible to separate the Greek discovery of irrational numbers from the unexcelled sense of Greek tragedy and poetry for the complexity and metaphoric structure of human relations and of man's place in the world. Conversely, it would be worth investigating what psychological, conceptual affinities there may be between certain failures in Greek political maturity and economic order and the inability of Greek mathematics to arrive at a genuine abstraction of space. The seventeenth-century duality of mathematics and philosophy, of mathematics and the arts, is crucial to modern civilizations. It was embodied in individuals such as Descartes, Kepler, Fermat, Pascal, Boyle and Leibniz. It permeated the confident rigour of philosophic and poetic language, the sense of a commanding at-homeness in natural and social experience which gives to the thought of a Galileo and of a Newton an unmistakable centrality. The music of the baroque, Vauban's military architecture, the mechanics of Huygens and the Bernouillis, exhibit a parallel stylishness, an economy of line and tension, which are functional in the general and in the algebraic sense. Today also, there are exceptionally gifted individuals in whom a mathematical and a humanistic sensibility are congruent. There is a subtle current of mathematical education and continued interest in some of the most interesting of modern literature: in novelists such as Musil and Broch, in the aesthetics of Valéry, in the poetry of William Empson, in the fantasies and "permutations" of Raymond Queneau. There is an explicit mathematical strain in certain schools of contemporary, totally programmed music and in kinetic art. Symbolic logic and linguistic analysis draw on modern algebra and on set theory. But the harmonic strengths of interaction, of concordant creation which mark

the role of mathematics and of the arts in fifth-century Athens and seventeenth-century western Europe, have largely disappeared. Why should this be?

The common-sense answer is that mathematics have become so difficult that only the specialist can hope to grasp any but their rudiments. It is no longer possible to be both humanistically or generally and mathematically literate at the same time. There is a psychological puzzle here. When was mathematics "easy"? Was Apollonius on conic sections "less difficult" for his contemporaries to understand than were, say, Fermat on prime numbers or Dirichlet on the application of Fourier analysis to number theory? In other words: is "difficulty" not, in part at least, a cultural, almost aesthetic criterion with its own relativism and psychological history? "In part, at least" is probably the crucial factor. As mathematics progress, a very sharp increase in intrinsic difficulty does seem to occur. If I referred, a moment ago, to Fourier series, this was not entirely accidental. It is with the Fourier series, and with Poisson's combination of the Fourier series with the Fourier integrals, that analysis, which is in turn the foundation of modern mathematical reasoning, seems to undergo a quantum leap of difficulty. I would almost want to express the change in temporal coordinates: the time-span of attention needed to grasp an analytic proof for the so-called Gauss Reciprocity Law or the Kronecker integral which allows an analytical treatment of spaces of more than three dimensions, is longer, more severely abstractive from the facilities of ordinary random consciousness, than any called for by Euclid or even by Descartes's analytical geometry. Indeed, nothing in man's history is more fascinating than the capacity of the intellect to pose mathematical problems which are at first insoluble but whose postulates carry with them both the obligation and possibility of subsequent solution. The tree literally grows as the roots deepen into the unknown. But whatever the contingent and inherent reasons, the place of mathematics in our general culture has become one of specialized isolation. The consequences have, I believe, been more damaging than we usually realize. The sense of fragmentation in our understanding of natural and social phenomena, which is a cliché of current self-consciousness, may well have its source in the widening split between a mathematical and a humanistic sensibility. This may be so in a very simple, statistically-distributive sense: modern mathematics probably enlists intellectual and spiritual resources very close to the top of the human curve; by drawing them off into a remote, highly insulated sphere of performance and mutual stimulation, it leaves the rest of our creative, intellectual reserves significantly impoverished. There is no direct feedback. But the problem is also one of the more general climate of imagination, of the habits of feeling which underlie our political and social conduct. Mathematics is a sober, fiercely self-critical enterprise; but it has, since Plato, hobbled with eternity. It is a deeply individual, almost private obsession; but it is also inevitably collaborative and the intense competition is one that is harnessed to a common, transcendent pursuit. Mathematics demands a unique degree of consistency and verifiability; but uniquely also, these criteria will frequently coincide with the unmistakable, though very difficult to define, ideals of beauty, of perfected shape. However complex and particular in formal focus, mathematics is universal, its grammar belongs to no single race or *milieu*. Consider these contrastive but correlative forces of sobriety and philosophic height, of individuality and teamwork, of verification and beauty, of local style and universal access—and you will have a shorthand list of some of the energies and qualities of feeling most lacking in the present tenor of our private and political existence. It cannot be wholly a coincidence that a society so vulnerable to boredom, to every type of intellectual and political mendacity or simplification, so trapped in the insistent chaos of the moment, should also be a society which is, mathematically, largely illiterate. The intimation that a city without musicians and geometers (related guilds) is a poorer, more threatened place, is as old as Pythagoras.

What can be done?

It is all too obvious that my own remarks are those of a mathematical illiterate, that I am myself a product of a largely split educational tradition. Those present here tonight will have a much more acute, responsible view of the educational problems than any I can show. All I can offer therefore is anxious generality. But if my preceding remarks have any merit, it may be that mathematics, like certain other human pursuits, is too serious a matter to be left only to mathematicians or even to teachers of mathematics.

A great deal of effort and intelligence (the Guinness Awards are a case in point) has gone into the revision of the school syllabus in mathematics, into the study of better ways of teaching and organizing mathematical skills in class. But so far as England goes, the resulting improvements have not affected the real problem: which is the premature fission between the sciences and the arts, between mathematics and the humanities. O-level mathematics are notorious for their low standards, for the fact that one can get by on almost nothing but a bit of expedient drill. Yet it is at O-level, often even before, that a major proportion of young people in England bid a lasting goodbye to mathematics. In short, mathematical illiteracy is not only condoned by implicitly facilitated and, I would nearly say, encouraged by the absurd system of premature specialization. After the age of fourteen or fifteen, a man or woman in this country need never again come up against a serious mathematical concept. This is, surely, a gravely disturbing phenomenon, made more so by the contrast with the far more broadly-based educational requirements pursued in France, in Japan, in the Soviet Union. The damage is many-sided. Let me stress only one aspect. Those whose mathematical awareness has ceased with O-levels are, today, fairly well cut off from any but the most second-hand, mendaciously predigested view of much of modern philosophy, of modern economics, of the forms of statistical argument which penetrate more and more of our politics, psychology and social thought. They have only a minimal access to the rapid developments in current genetics and biology. They are cut off completely from anything but Sunday-newspaper physics or cosmology. Much of what is most vital in the modern condition of mind and of sensibility is wholly inaccessible to those who last drew a tangent or extracted an algebraic root in some loathed school-room in early adolescence. Tomorrow, the status of the mathematical illiterate may be worse. Computers, data-banks, retrieval systems of every description will probably bring on changes in daily economic and social life fully comparable to those caused by the first industrial revolution. A certain level of mathematical understanding, a certain unforced intimacy with the notion of algebraic series, of scatter, of fields, of classes, will be the key to this new world. Is anyone really educated who has not had mathematical instruction at least to the point where he leaves secondary schooling?

Of course there are obstacles, foremost among them the universities with their acceptance formulas based on an absurdly narrow set of A-levels, with their unwillingness—until very recently—to allow a man to continue working on parallel or interdisciplinary fronts. But there is also, I feel, widespread doubt as to the “teachability” of mathematics to those who seem recalcitrant or who plead some inherent inadequacy in their own intellectual equipment. Here there is clearly a very complex mixture of cultural, psychological and social factors. All I am able to voice is the conviction that mathematics could be taught well and taught longer to every normal schoolboy or schoolgirl if it was conceived of and presented as one of the very great languages of man. This, of course, can be meant in a technical way. Different periods in this history of mathematics, different individual mathematicians have their recognizable style and “dialect” (a page in a paper by Hilbert or Poincaré is, stylistically, visually, as unmistakable to a trained eye as would be, say, a page in a Bach score). But I intend “language” in the widest sense:

mathematics, like verbal speech, is an organic, historically formed, way of perceiving, of stating and of making predictions about reality—either external reality or the no less substantive reality of thought. It is a way, incomparably subtle, coherent and often witty, of recording, elucidating and translating experience. Verbal speech, music and mathematics are very likely the three principal modes of utterance of human civilization, the three signal codes of the mind when it is most fully in play (as distinct from the more “bodily” communicative codes of gesture and the plastic arts). Seen as a major way of “speaking about”, of “declaring” our relations to the world, mathematics cannot be relegated to the status of a rebarbative speciality to be dropped from the curriculum with indecent haste. It must be viewed as an integral element of human articulation, to be mastered as one masters one’s native speech, at least to a certain modest level of fluency.

A good many heuristic devices follow on such a view. Concepts and problems can be taught genetically, according to the often dramatic logic of their actual historical development. The history of the liberation of space from visual, tactile limitations related Euclid to Descartes, Descartes to Riemann. The long struggle to prove that every integer can be represented as a finite product of prime numbers in which some factors may repeat themselves and that the representation is unique, leads from the ninth book of Euclid’s *Elements* in 300 BC to Gauss’s *Disquisitiones arithmeticae* of 1801. Why did this fundamental theorem elude the best mathematicians for 2,100 years? Introduced to the problem in its historical setting, the student will not only pick up some very handsome arithmetic but a degree of insight, difficult to structure as lucidly in any other discipline, into the growth and limitations of the life of the mind in western culture. Why not introduce a schoolchild to the notion of function as it actually evolved from the first years of the seventeenth century, to the eighteenth-century understanding of trigonometric functions and the nineteenth-century realization of boundary values? Again, the history is one of intellectual adventure, temporary defeat and the forward leap of tensed imaginative energies, and it is a history rich in general implications. Wherever possible, indeed, mathematics ought to be seen and taught as part of the structure of the relevant period, with its correlates in art, music, philosophy and the social climate of the age. The deep, genuine parallels with the best teaching of language and of literature should be evident. It follows also that those who teach mathematics to schoolchildren should be among the most highly trained, generally literate and imaginatively excited in our community. Their prestige and financial status should fully match these exacting needs. This is the case in Russia, in the best German and Swiss *Gymnasias*, in the *grands lycées* of France. Is it the case in England? How can a discipline prosper when so many who teach it to human beings at the crucial stage of mental development feel themselves to be among the defeated?

It would be impertinent for a layman to say more. And it has all been said so very much better by a young man not yet twenty-one who in the last night of his life established the basis of modern algebraic theory and topology. Galois’s famous letter-memoir of May 29, 1832, concentrates every aspect of the passionate stress and adventure of mathematics. There is the exultation, so characteristic of mathematical feeling, at the fact that the imagination can glimpse a great field about which “at present we know nothing”. There is the grim note of individual limitation, familiar even to those mathematicians who have known a full career: “I have not time enough and my ideas about this immense terrain are not yet well developed.” There is, above all else, the possession of a living being by an energy, by a delight of pure thought and rigour so intense as to overcome even the proximity of death. How diminished is a literacy which cannot include such a text among its classics.

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Cores of the Terrestrial Planets

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The compositions of the cores of the terrestrial planets have been re-examined following a calculation by O. G. Soroktin that the iron oxide Fe_2O is stable at pressures reached in the Earth's core and his suggestion that the outer core may consist of Fe_2O . It is shown that the idea of an Fe_2O outer core in the Earth can be fairly well reconciled with a common overall composition for the planets Earth, Venus and Mars in a way that avoids the main objections to the earlier phase-transition theory.

THE presently favoured theory that the Earth's outer core consists predominantly of iron (possibly alloyed) requires^{1,2} the overall compositions of the planets Earth, Venus and Mars to be markedly different. It is difficult to reconcile this result with accretion theories of the origin of the planets¹⁻³.

In an early attempt at reconciliation, Ramsey⁴ and myself⁵ independently advanced a "phase-transition theory" which treated the Earth's outer core as a high-pressure modification of the lower mantle material. This theory agreed with a common overall composition for the three planets and fitted well the then available observational data on the masses and radii of Venus and Mars and the polar flattening of Mars. The theory later met with difficulties.

Revisions of the planetary observational data reduced the quality of the original fit. The most serious surviving discrepancy is that the preferred observational estimate⁶ of the radius of Venus is now $6,052 \pm 10$ km, as against 6,270 km predicted⁷ on the phase-transition theory.

The original Earth models *A* and *B* (refs 8, 9) used in deriving models of the other planets have had to be amended (see refs 10, 11) to allow (among other things) for the reduction of the estimated Earth's central density¹² from about 18 to 13 g cm⁻³, and of the estimated moment of inertia coefficient¹³ for the Earth from 0.3336 to 0.3309. Both revisions have slightly worsened the fit with Venus and Mars.

Geochemists have found difficulty, principally on the question of the packing of oxygen atoms in the lower mantle material, in reconciling the proposed phase transition with the large density jump (in the ratio 0.7 or more) at the Earth's mantle-core boundary, *N* say.

Transitory shock-wave experiments carried out in laboratories at high pressures have failed to supply positive evidence that a phase transition occurs at *N*.

Alternative Model

Having regard to various uncertainties and questions of experimental interpretation, I think it is unsound to assert,

as do some investigators, that the difficulties have "disproved" the theory—indeed the theory still has some notable followers. But the case against it is sufficiently strong to make it desirable to seek alternatives.

As a possible alternative, I propose a theory which avoids the principal difficulties of the phase-transition theory while retaining the important feature that the pressure p_c at *N* is critically involved in the changes of property at *N*. The theory incorporates a suggestion of O. E. Soroktin (personal communication) that the Earth's outer core consists of Fe_2O . Soroktin has calculated that this oxide, which is unstable at ordinary pressures, becomes stable at the pressures in the Earth's core and has a density-pressure relation matching that in the Earth's outer core.

In other respects, I have deviated substantially from Soroktin. Whereas he attributes the occurrence of Fe_2O in the outer core to a breakdown of FeO into Fe_2O and oxygen, I have associated the occurrence of the Fe_2O with the equation $\text{Fe}_2\text{O} \rightleftharpoons \text{FeO} + \text{Fe}$, my reason being the extent of planetary fit that can be thereby achieved.

My proposal, which I refer to as the " Fe_2O theory", envisages a model family of planets with the following properties:

All planets of the family are composed of two primary materials—a basic mantle material *X*, and Fe_2O . (This is subject to qualification below.) For present purposes, the composition of *X* need not be specified, but *X* is likely to include some FeO .

In all the planets, the ratio of the mass of *X* to the mass *M* of the planet is the same.

In those planets which contain Fe_2O , the Fe_2O occurs as a distinct zone (the "outer" core) throughout which $p \geq p_c$, where p is the pressure.

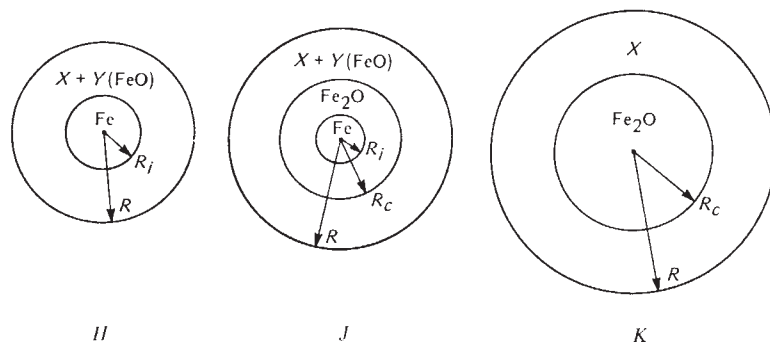
In those planets where the first two properties would entail $p < p_c$ in an Fe_2O zone, some or all of the Fe_2O has broken down into FeO and Fe . This FeO , which I refer to as *Y*, forms part of the mantle and is additional to any FeO that may be part of *X*. The Fe falls to form an "inner" core.

Details of the Model

The second property secures a common overall composition for the family. The masses of *Y* and Fe in any planet are such as would combine precisely to form Fe_2O when $p \geq p_c$, and the ratio of the total mass of Fe , Fe_2O and *Y* to *M* is the same for the whole family.

The family has three subsets—*H*, *J*, *K*, say (Fig. 1). The subset *H* includes the smallest planets which have no Fe_2O zones and thus no outer cores; they have mantles composed of *X* and *Y* mixed, and inner cores of Fe . The subset *K* includes the largest planets which have mantles composed purely of *X*, outer cores of Fe_2O , and no inner cores. The subset *J* consists of intermediate planets which have mantles composed of *X* and some *Y*, outer cores of Fe_2O , and inner cores of Fe . For all the *J* planets, $\rho = \rho_c$ at the top of the outer core. The core-mantle mass ratios are the same for all *H* planets, and for all *K* planets (but the ratio is different for *H* and *K*).

Fig. 1 Materials in the interiors of the three subsets (*H*, *J*, *K*) of terrestrial planets. The outermost zones, all containing the material *X*, are mantles. The Fe_2O zones are referred to as "outer cores", the Fe zones as "inner cores".



Earth and Venus would correspond to members of *J*, Mars of *H*. Unfortunately, no known planets correspond to *K*, so that an observational test of the subset *K* is not available. From evidence on the Earth, all Fe_2O zones (outer cores) would be expected to be fluid, and Fe zones (inner cores) solid.

The Fe_2O theory resembles the phase-transition theory in that all *J* planets have the same (critical) pressure p_c at the mantle-core boundaries, and all *H* planets lack outer cores. To a considerable extent, it is this feature which enables both theories to go close to fitting a common overall composition for Earth, Venus and Mars. Differences in detail arise because, in planets smaller than Earth, the Fe_2O theory entails larger iron (inner) cores than does the phase-transition theory.

Agreement with Observation

I have started from a simplified Earth model, taken to be a member of the subset *J*, in which the mean densities of the mantle, outer and inner cores are 4.5, 11 and 13 g cm^{-3} , respectively. I have provisionally neglected variations of density inside planetary zones and taken the ratio of the mean densities of *X* and *Y* as 0.7. For a first approximation, differences of compression between corresponding zones of the *J* planets have also been neglected. Possible temperature differences between different planetary interiors, volume changes due to chemical interaction, and so on, in the mixing of *X* and *Y* in mantles, and the presence of nickel in cores have been neglected as likely to affect the essential numerical detail only slightly.

With these simplifications, the postulates formally determine numerical details for all the *J* planets. In particular, a set of values of *M*, *R* (radius of the planet), R_c (radius of the outer core) and R_i (radius of the inner core) is determined.

The observational mass of Venus¹⁴ is 4.87×10^{24} kg. For the *J* planet with this mass, the simplified Fe_2O theory gives $R=6,010$ km, $R_c=2,710$ km, $R_i=2,110$ km. Allowance for the neglected compression differences will increase this estimate of *R* because Venus, being smaller, is less compressed than the Earth. Thus the Fe_2O theory fits the observed radius ($\sim 6,050$ km) of Venus as well as can be expected.

The value 600 km yielded for the thickness, *T* say, of the (presumed fluid) Venus outer core is also interesting. It may be contrasted with the value exceeding 900 km (ref. 7) on the phase-transition theory and substantially larger values on theories which assume a predominantly iron outer core. By yielding the lowest value of *T*, the Fe_2O theory accounts best for the failure to observe a significant magnetic field around Venus; the presumed seat of the Earth's magnetic field (the Earth's outer core) is 2,200 km thick.

The postulates also determine the ratio of the mass of the inner core to the mass *M* of any *H* planet as approximately 0.15. For a particular *H* planet of assigned mass, a first approximation to *R* is then formally yielded if values are available for the mean densities, ρ_1 and ρ_2 say, of the core and mantle. For Mars, $M=0.642 \times 10^{24}$ kg. For the *H*

planet with this value of *M*, taking $\rho_1, \rho_2=9.0, 3.7 \text{ g cm}^{-3}$, the simplified Fe_2O theory gives $R=3,354$ km; the yielded value of *R* is increased by 10 km if the trial value of ρ_1 is decreased by about 1.0 g cm^{-3} , or of ρ_2 by about 0.03 g cm^{-3} . Thus uncertainties in the appropriate values of ρ_1 and ρ_2 could possibly permit raising *R* to about 3,370 km, which is, however, less than the presently preferred observational value^{3,15,16} of $3,388 \pm 5$ km.

When hydrostatic conditions are assumed throughout Mars and ρ_1 and ρ_2 are treated as constant, the Fe_2O theory yields $f=1/191$ and $\delta=18$ km, where *f* and δ are, respectively, the polar flattening and the excess of the equatorial over the polar radius. These results agree closely with the observed *f* and δ (see refs 3, 17). But allowance for density variation inside the mantle is likely to lower the calculated values to 1/200 (or even less) and 17 km. These last values are on the border¹⁷ of the limits of error of the hydrostatic theory.

The phase-transition theory gives $R=3,390$ km and $f \approx 1/188$ (ref. 18), assuming hydrostatic conditions. Thus the phase-transition theory gives the better fit with Mars unless the departures from hydrostatic conditions are considerable. This is principally because the Fe_2O theory requires a larger core, radius $R_i \sim 1,400$ km. It is likely that seismic records taken on Mars will in due course provide an observational estimate of R_i and so serve as a further test of the theories. (The gathering of seismic records on Venus is likely to be more difficult.)

Ironically, after the phase-transition theory had been shown to fit closely the then assumed observational estimate $R=3,390$ km for Mars, several investigators were disposed to reject that theory by insisting on observational values of 3,350 km or less¹⁷. A value of 3,350 km would fit the Fe_2O theory extremely well.

Assuming, as usual, that the mantle and iron core of Mars are solid (ref. 19) the Fe_2O theory, in common with the phase-transition theory, accounts satisfactorily for the failure to detect a magnetic field around Mars.

So the provisional calculations indicate that the Fe_2O theory fits the observational *M* and *R* for Venus extremely well, and *M*, *R* and *f* for Mars moderately well. The theory also gives the closest agreement of all theories with the observational evidence on magnetic fields.

Like all theories that are not extremely *ad hoc*, the Fe_2O theory does not fit the planet Mercury as Mercury now is, although the theory has an advantage over the phase-transition theory in requiring a substantially larger iron core. As I have previously suggested²⁰, Mercury, through its proximity to the Sun, has probably lost much of its primordial mantle by volatilization. The Fe_2O theory would suggest that the primordial Mercury (assumed to belong to the subset *H*) was larger than Mars.

The low density of the Moon precludes it from belonging to the family of planets considered here. The *H* planet with the Moon's mass would have an iron core of radius about 700 km, which is appreciably too large to fit evidence on the Moon's mean density and moment of inertia. Thus

the Fe_2O theory here merely confirms that the Moon must have had an exceptional origin.

I hope to present later calculations which take detailed account of compression and density variation inside the planetary zones.

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Skull of *Hesperornis* and Early Evolution of Birds

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Additional preparation of a specimen found a century ago permits complete reconstruction of the skull of the Cretaceous bird *Hesperornis*. This skull provides new evidence on the evolution of the avian palate and the origin of avian cranial kinesis, and further substantiates the evolution of birds from theropod dinosaurs.

In spite of some disagreement about the immediate ancestry of birds¹⁻³, there is little doubt that the Late Jurassic *Archaeopteryx lithographica* is the earliest known representative of the class Aves. *Archaeopteryx* was a very primitive reptile-like bird—as Ostrom³ states, the *Archaeopteryx* specimens would probably have been identified as dinosaurs if feather impressions had not been preserved with them. By the end of the Cretaceous, about 70 m.y. after *Archaeopteryx*, the avian fauna apparently consisted exclusively of birds of modern aspect referable to Tertiary orders⁴. The only well preserved specimens intermediate in age and morphology between the dinosaur-like *Archaeopteryx* and modern birds are two genera, *Hesperornis* and *Ichthyornis*, described by O. C. Marsh a century ago from rocks of early Late Cretaceous age.

During Marsh's expedition in 1871 to the marine Niobrara Chalk of western Kansas, he collected much of the post-cranial skeleton of a large swimming bird, which he named *Hesperornis regalis*⁵. Shortly thereafter a complete skeleton of *H. regalis* (Yale Peabody Museum 1206) was found. The skull of this specimen included the crushed braincase and pieces of the upper and lower jaws. Many of these bones were cleaned on one side only and never removed from the original matrix. The braincase and other cranial fragments of YPM 1206, together with a broken but

uncrushed occipital segment of another braincase (YPM 1207), formed the basis for the discussion and reconstruction of the skull of *Hesperornis* given by Marsh in his monograph on the Odontornithes⁶, or toothed birds. Marsh's skeletal reconstruction shows *Hesperornis* to have been a flightless, predaceous bird, probably similar in habits and diet to the living loons.

Following the publication of Marsh's monograph, Lucas⁷ described another partial skull of *Hesperornis regalis*, including the premaxillae and parts of the mandibles (US National Museum 4978), and a nearly complete but crushed skull of *Hesperornis gracilis* (University of Kansas 2287). *H. gracilis* differs from *H. regalis* only in being slightly smaller.

Marsh stated in his monograph⁶ that "in its more important characters, the skull of *Hesperornis* resembles that of the Ratitae, or Struthious birds". This view, as is discussed later, was immediately challenged by D'Arcy Thompson, and further discredited by Lucas, Shufeldt and Heilmann, none of whom restudied Marsh's original material. A separate, more recent controversy concerns the presence of teeth in the skulls of *Ichthyornis*⁸ and *Hesperornis*⁹. A new partial mandible of *Ichthyornis* and restudy of the best preserved Yale skull of *Hesperornis* (YPM 1206) has confirmed Marsh's descriptions. The jaws of both *Ichthyornis* and *Hesperornis* bore true thecodont dentitions¹⁰.

The retention of thecodont teeth in birds that appear in many other features to be fully modern is evidence that *Hesperornis* and *Ichthyornis* are truly intermediate between *Archaeopteryx* and modern birds. *Hesperornis*, being represented by parts of four skulls, is thus of great importance in evaluating several problems of early avian evolution, including the early evolution of the palate and the origin of avian cranial kinesis. The origin and function of the intramandibular articulation of *Hesperornis* is also discussed later.

Structure of the Palate

The new reconstruction of the skull of *Hesperornis regalis*, shown in Fig. 1, incorporates the information gained

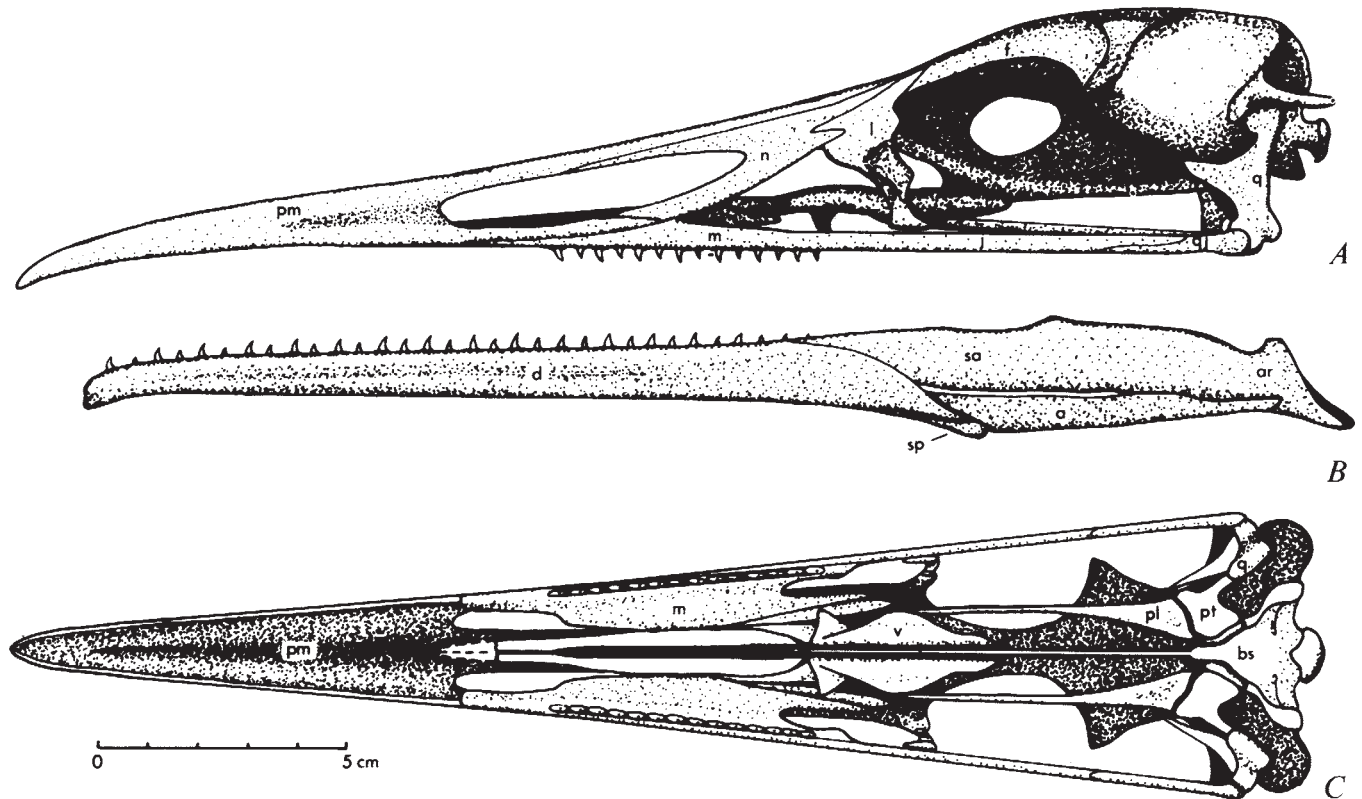


Fig. 1 New reconstruction of the skull of *Hesperornis regalis*. Cranium (A) and mandible (B) in lateral view, palate (C) in ventral view. a, Angular; ar, articular; bs, basisphenoid; d, dentary; f, frontal; j, jugal; l, lacrimal; m, maxilla; n, nasal; pl, palatine; pm, premaxilla; pt, pterygoid; q, quadrate; sa, surangular; sp, splenial; v, vomer.

from further preparation of the skull of YPM 1206, and from detailed comparison of it with the other three known skulls of *Hesperornis*. One of the most important discoveries resulting from preparation of YPM 1206 is the true identity of several of the bones of the palate. The bone illustrated by Marsh as a vomer (ref. 6, Plate 2, Fig. 8) is in fact an almost complete left palatine, preserving articular facets for both the pterygoid and the vomer. The bone identified in Marsh's Plate 2, Fig. 9 as a palatine is actually the right vomer.

The corrected relationship of the bones of the palate of *Hesperornis* is illustrated in Fig. 1C. The pterygoid is a relatively small bone with a concave articular surface contacting the quadrate just above its medial keel. The medial process of the pterygoid terminates in a large facet which articulated with a short, broad "basipterygoid" process of the basisphenoid. The pterygoid and palatine were joined along a relatively immovable joint. The palatine is broad at its posterior end, and tapers anteriorly. It is fused anteriorly to the dorsolateral surface of the posterior part of the vomer. The vomers are long thin bones which are expanded posteriorly in association with the openings of the internal nares. They articulated with a narrow parasphenoid rostrum posteriorly, and apparently formed an anterior extension of it. The vomers were separate for all of their preserved length but the anterior ends are missing and may have been fused. It is not possible to determine exactly the original conformation of the expanded posterior end of the vomers, again because of breakage. The vomers certainly did not approach contact with the pterygoids. Approximately the medial third of each vomer was sutured to the corresponding maxilla.

All the palatal bones are now disarticulated but, considering the morphology of the articular surfaces, it is unlikely that significant relative movement was possible within the palatine-vomer-maxilla complex. The palatine and

pterygoid were firmly joined, though not fused. The articular surfaces joining the quadrate and pterygoid, and the pterygoid and basisphenoid are smooth and some movement between these elements no doubt took place.

Huxley, in his 1867 classification¹¹, separated the ratites (in his opinion a primitive group) from other birds, citing their unkeeled sternum, lack of a sphenoid rostrum-palatine articulation, strong "basipterygoid" processes arising from the basisphenoid, undivided proximal head of the quadrate and so forth. Marsh⁶ based his statement of the ratite affinities of *Hesperornis* chiefly on these four characters. D'Arcy Thompson¹² challenged Marsh's interpretation, and from a study of Marsh's figures concluded that *Hesperornis* was related most closely to the living loons and grebes. Lucas⁷ supplemented Thompson's list of differences between *Hesperornis* and the ratites. Finally, Shufeldt¹³ published a reconstruction of the palate of *Hesperornis* based on Marsh's published figures, several poor photographs of the specimens, and several skulls of loons—not surprisingly, his reconstructed palate closely resembles the palate of a loon. Little was left for Heilmann¹⁴ to do in his classic book on the origin of birds, but to quote Shufeldt¹³: "It will once and for all disabuse the mind of the paleontologist with respect to there being any agreement of characters when we come to compare the skull of the ostrich with that part of the skeleton of *Hesperornis*". As Marsh's specimens were never restudied, this view has gone unchallenged.

More recently De Beer¹⁵ reasoned that because the plumage and the lack of fusion of the bones of the braincase in ratites are derived by neoteny, and as the palate of *Archaeopteryx* was neognathous (based on the reconstruction of Kleinschmidt¹⁶), it is probable that the "palaeognathous" palate of the living ratites is derived by neoteny from the "neognathous" type characteristic of the majority of living birds. Unfortunately Kleinschmidt's recon-

struction of the palate of *Archaeopteryx* is completely hypothetical—the morphology of the palate cannot be determined in the known specimens. In spite of this, De Beer's idea that the palaeognathous avian palate is derived from a neognathous ancestor has since been elaborated¹⁷, and has apparently gained wide acceptance¹⁸.

The structure of the palate of *Hesperornis* is obviously of primary importance in considering the evolution of the avian palate. When Pycraft¹⁹ originally proposed a fundamental dichotomy between the palaeognathous and the neognathous birds, he stated that the skull of the Palaeognathae differs from all other known birds in having a firm, immovable pterygoid-palatine connexion. Huxley's diagnosis¹¹ of the dromaeognathous palate (=palaeognathous palate of Pycraft) based on the absence of a palatine-sphenoid contact and on the presence of basiptyergoid processes arising from the body of the basisphenoid, also distinguishes the palaeognathous palate from that of neognathous birds.

The palate of *Hesperornis* (Fig. 1C) was clearly palaeognathous in all diagnostic features. The pterygoid and palatine were firmly joined. The palatines were separated from the sphenoid rostrum by the pterygoids, and strong basiptyergoid processes (which articulated with the pterygoids) were present. Considering the morphology of the palate of *Hesperornis*, the palaeognathous condition is probably truly primitive in birds. This does not, of course, imply that the ratites are descended from flightless ancestors as some early writers maintained. There is good anatomical evidence that ratites evolved from ancestors capable of flight¹⁵.

Recent studies on the egg white proteins of ratites^{20,21} indicate that the large ratites (*Struthio*, *Casuaris*, *Dromaius* and *Rhea*) are more closely related to one another than to any other group of living birds, supporting classification of them together in the order Struthioniformes. The kiwis (*Apteryx*) and the tinamous apparently constitute two separate groups, and are retained in the orders Apterygiformes and Tinamiformes respectively. These three living groups of palaeognathous birds seem to represent three surviving orders of the first major radiation of birds. Most of this Cretaceous radiation (which included *Hesperornis*) is now extinct. To quote Huxley¹¹: "Hence, in all probability, the existing Ratitae are but the waifs and strays of what was once a very large and important group."

Intracranial Mobility

Most birds in addition to raising and lowering the lower bill, are able to raise and lower the upper bill relative to the braincase. The general function of this cranial "kinesis" presents one of the most interesting unsolved problems of avian skull morphology. Simonetta²² and Bock²³ have published important reviews of the kinetic movement of birds, and Frazzetta²⁴ has reviewed the cranial kinesis of lizards. Raising the upper bill by bending the nasals and premaxillae (as in living palaeognathous birds) is termed rhynchokinesis. Raising the upper bill about a transverse hinge between the frontals and premaxillae (as in neognathous birds) is termed prokinesis. Mesokinesis is the condition in some reptiles where the upper jaw is raised by means of a transverse hinge between the frontal and parietal bones.

In *Hesperornis* the premaxillae and nasals overlapped the frontals by about 1.5 cm. The premaxillae and nasals appear to have been firmly sutured to the frontals, ruling out a prokinetic hinge at this point. The premaxillae and nasals were joined above and below the external nares, forming a single rigid member and eliminating rhynchokinesis. The quadrates, however, appear to have been moveable (streptostylic), a characteristic which in modern

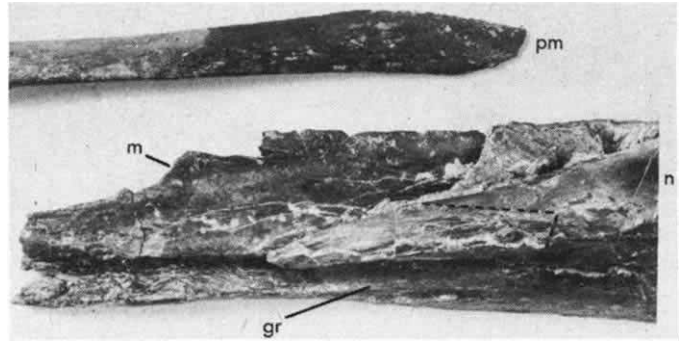


Fig. 2 Dorsal view of left maxilla (m) of *Hesperornis regalis*, YPM 1206, showing groove (gr) which fits over nasal-premaxilla bar. Nasal (n) preserved against maxilla. Nasal and premaxilla (pm) in lateral view, dashed line shows degree of overlap of premaxilla on nasal.

birds is associated with some kind of kinetic mobility. The articulation of the maxillae with the nasals and premaxillae reveals how streptostyly was associated with an akinetic skull in *Hesperornis*.

In the Yale skull of *Hesperornis* (YPM 1206) the fused premaxillae, the left and right nasals, and the left and right maxillae are all disarticulated, permitting examination of all articular surfaces. Rod-like ventral processes of the premaxillae and nasals were joined by a long overlapping suture below the external nares (Fig. 2). Together these formed a smooth subnarial bar on each side of the skull. A smooth groove is present above the tooth row on the dorsal surface of each maxilla (Fig. 2). The corresponding subnarial bar clearly fits into this groove. The smooth surfaces of the maxillary grooves and subnarial bars indicate that they were not firmly joined but articulated by a sliding mechanism, with the subnarial bars of the nasals and premaxillae forming rails to guide forward and backward movements of the maxillae.

Further evidence that the maxillae articulated freely with the nasals and premaxillae is provided by the degree of disarticulation of the skull of *Hesperornis gracilis* (UK 2287). This skull is crushed, and is now preserved palate upward on a slab of chalk. The subnarial bars formed by the nasals and premaxillae have remained essentially intact on both sides of the palate. The maxillae have both separated cleanly from the subnarial bars, and they are either crushed into the centre of the palate or missing. It seems unlikely that both maxillae would be lost from an otherwise articulated skull if they had been firmly joined to it.

Thus, streptostyly in *Hesperornis* apparently was associated only with fore-aft movement of the maxillae relative to the rest of the skull, and not with prokinesis or rhynchokinesis as it is in modern birds. The exact distance that the maxillae were able to slide relative to the rest of the cranium cannot be determined accurately, but it seems to have been approximately equal to or slightly greater than the distance between two adjacent teeth, that is, about 2 to 3 mm. The function of this sliding mechanism is not clear. It possibly assisted in pulling prey into the mouth, or in disengaging the maxillary teeth from prey. In this regard it is significant that the upper teeth occur only in the sliding maxillary segments. The premaxillary part of the upper bill was edentulous and apparently covered with a horny bill similar to that of living birds.

The absence of a standard form of kinesis in *Hesperornis*, a primitive bird because of its geological age and its retention of thecodont teeth, requires a new interpretation of the evolution of kinesis in birds. In his review of avian kinesis Bock²³ attempted, as Versluys²⁵ had previously, to trace kinesis continuously from a kinetic reptilian ancestor to birds. Bock reasoned that because none of the biological roles generally ascribed to avian kinesis imparts any obvious advantage, kinesis was probably inherited from

reptilian ancestors rather than developed independently in birds. Following this reasoning, Bock interpreted the poorly preserved skull of *Archaeopteryx* as mesokinetetic.

Two recent developments argue against this. As described earlier, the skull of *Hesperornis* was akinetic. Moreover, Ostrom's³ evidence of direct phyletic relationship between *Archaeopteryx* and coelurosaurs indicates that birds evolved from this group of theropod dinosaurs. It has not been convincingly demonstrated that any theropod possessed a kinetic skull. Colbert and Russell²⁶ interpreted the skull of *Dromaeosaurus albertensis* as mesokinetetic, but the supposed mesokinetetic hinge is not preserved in the only known skull of this species. Furthermore, the quadrate and pterygoid on each side of the skull of *Dromaeosaurus* have broad, thin, overlapping processes which appear to have been joined together tightly. The suggestion that the skull of *Deinonychus* may have been kinetic²⁷ was based only on the disarticulated nature of the preserved specimens. Restudy of *Dromaeosaurus* and *Deinonychus* has provided no evidence that either had a kinetic skull. Russell²⁸ demonstrated that the skulls of the large theropods (tyrannosaurs) were akinetic. Consequently it seems very unlikely that birds inherited any form of kinesis from their reptilian ancestors. Even if theropods and *Archaeopteryx* had mesokinetetic skulls, this would not explain the origin of avian rhynchokinesis and prokinesis. Cranial kinesis in birds seems to have had an independent evolutionary origin.

Unfortunately the general function of avian kinesis is still not well understood. The discovery that the toothed bird *Hesperornis* was akinetic suggests a new explanation of the function of kinesis in birds. Teeth such as those of *Hesperornis* are useful for holding, and for puncturing and ripping prey. The mandibles of *Hesperornis* articulated with the quadrates by a sliding joint permitting fore and aft motion of the mandibles relative to the cranium. By coordinated movement of the mandibles and maxillae food could be advanced into the mouth. The function of the bill in most birds lacking teeth must be to slice rather than to puncture. The slicing ability of the upper and lower bills of modern birds is related not only to the sharpness of their edges, but also to their angle of relative movement. As the lower beak moves almost parallel to the upper, they form an efficient slicing device. It is probable that the conventional forms of avian kinesis evolved after the loss of teeth and the development of a slicing horny bill.

Kinesis increases the amount of possible movement of the lower bill relative to the upper. In the pheasant *Phasianus colchicus* (of skull length 73 mm) the total possible fore-aft motion of the articular bone relative to the quadrate is 2 mm. Thus, if the pheasant skull was akinetic, the fore-aft motion of the lower bill relative to the upper bill would be limited to 2 mm. The skull, however, is kinetic. If the upper bill is depressed when the lower bill is protruded, and then raised by pressure of the lower bill as it is adducted, the lower bill moves approximately 6.5 mm relative to the upper (determined by manipulation of a freshly killed specimen). This is more than a three-fold increase in the relative movement of the two elements. Thus a kinetic skull greatly increases the relative movement of the upper and lower bills, and so significantly augments shearing with the beak. Both the mandibular adductor and the pterygoideus muscles in most birds are aligned with the shearing region of the beak, suggesting that the beak can be used efficiently for shearing during both adduction and protrusion of the lower bill²⁹.

The evolution of prokinesis and rhynchokinesis from an ancestor with a skull similar to *Hesperornis* involved refusion of the maxillae to the premaxillae and nasals, and either the development of a line of bending between the frontals and the premaxilla-nasal rostrum, or the reduction and loss of the postnasal connexion between the nasals and maxillae. Prokinesis and rhynchokinesis may have

evolved independently in neognathous and palaeognathous birds, or rhynchokinesis may have evolved from some form of prokinesis. The exact evolutionary sequence cannot be determined without additional well preserved skulls of fossil birds.

Intramandibular Articulation

One additional feature of *Hesperornis* should be mentioned. Each mandible of *Hesperornis* is composed of six bones arranged in approximately the same configuration as those of most theropod dinosaurs. The external mandibular fenestra lies above the angular and below the surangular (Fig. 1B) as is typical of reptiles. Thus Cracraft's³⁰ speculation about the avian rather than reptilian position of this fenestra in the poorly preserved mandible of *Archaeopteryx* is probably incorrect.

There is an articulation between the splenial and angular in each mandible of *Hesperornis* (and *Ichthyornis*). Gregory³¹ drew attention to this intramandibular joint and to a similar one which apparently evolved independently in mosasaurs. It has not been noted previously that the mandibles of many theropod dinosaurs (such as *Deinonychus* and *Dromaeosaurus*) have an intramandibular articulation identical to that of *Hesperornis*. The intramandibular articulation in *Hesperornis* may thus be an inherited character rather than one which evolved independently, as Gregory thought, in connexion with fish predation. The function of the intramandibular articulation in *Hesperornis* and theropods was probably to permit medial rotation of the posterior segment of the mandible independently of the dentary, thus facilitating disengaging the lower teeth from prey as the mandible was protruded. The presence of an intramandibular articulation in *Hesperornis* seems to be an important additional character linking early birds to a theropod ancestry. Unfortunately the mandible of *Archaeopteryx* is too badly damaged to determine if an intramandibular articulation was present in that form.

I have profited greatly throughout this study from discussions with Professor J. H. Ostrom, Yale University. Others who have assisted will be acknowledged in a more detailed paper, in preparation, on the skull of *Hesperornis*.

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LETTERS TO NATURE

PHYSICAL SCIENCES

Tidal Tilt Measurement in Europe

FOR many years instruments have been installed in underground observatories throughout Europe and elsewhere to observe the tilting of the Earth's surface caused by Earth tides and the loading effects of the ocean and shallow-sea tides. Analysis methods have been developed to permit comparison between the measured data and theoretical tides generated from astronomical data and information concerning Earth rigidity and the distribution of tidal waters. But the agreement between theoretical and observed tides has been poor and is often inconsistent between adjacent observatories, especially in the relatively dense European network.

In some cases where several instruments are installed in different parts of the same observatory¹ discrepancies between identical instruments have been observed. The discrepancies consist of both variations in amplitude and phase of the observed tidal harmonics and although it is possible to account for incorrect calibration as a source of amplitude errors, phase differences are not to be expected from the instruments themselves.

We discuss here a possible way by which amplitude and phase errors occur in tilt measurement, and we introduce the concept of "site generated tilting" to account for the tilt inconsistencies found within underground observatories.

The tidal magnitude at a site may be described using a spectral function $H_T(f)$ which is due to the local site tilt $S_T(f)$, the tilt due to the Earth body tide $G_T(f)$ and the ocean and sea loading tide $L_T(f)$ such that

$$H_T(f) = S_T(f) + G_T(f) + L_T(f) \quad (1)$$

The functions are complex, having both amplitude and phase. Thus the addition is a vector addition.

The differences between the observed and theoretical tilts determined where no allowance is made for the site effects are quoted by Lennon and Baker² to be $\pm 40\%$ in magnitude for the North-South M_2 tilt and $\pm 15\%$ for the East-West M_2 tilt. Phase errors of $\pm 10\%$ are present in both.

The site effect vector necessary to cause these effects therefore must be of the same order of magnitude as both the body-Earth tilt and the sea and ocean load tilts themselves and it must in general differ from both the other effects in phase. It is not possible to attribute the observed phase anomalies to the instruments. Also, no likely site structure will affect the phase of local tilting and it is difficult to think of a plausible structure which will significantly affect the amplitude.

It has been suggested that anomalies in the diurnal O_1 and K_1 tides are due to daily climatic effects. Our experience with strainmeter measurements suggests that the thermal noise is small in reasonable sites. In any case, in the frequency transform, climatic effects can be clearly distinguished from tidal peaks.

The only source of local tilting with the right sort of spectrum is to be found in the rock stresses set up by tides. These stresses are determined by the tidal strain field. Where inhomogeneities of rock properties occur, local anomalies in the strain fields result. Associated with these anomalies are tilts. The

magnitude and phase of the tilts are dependent on the magnitude and phase of the components of the tidal strain tensor for the region and on the geometry of the rigidity inhomogeneity.

Not all the components of the body strain tide are in phase with the tidal tilt components; for example, the East-West linear strain is 90° out of phase with East-West tilt, but North-South tilts and strains are in phase. This applies to both the diurnal and semidiurnal tidal species. Phase differences may also occur for tidal tilts and strains due to tidal loading but a discussion of this would be elaborate.

As an example of the way in which tilt-strain coupling may occur we consider the tilts induced in a tunnel wall due to a linear strain field imposed at a distance from the tunnel. This problem is relevant to the manner in which pendulum type tiltmeters are sited in observatories at present.

The problem of tunnels in elastic media subject to various stress fields is covered extensively in the literature of rock mechanics. The change of radius of a tunnel in an infinite block subject to a strain in regions distant from the tunnel is given by³

$$\delta r = e_{xx} r_0 (1 + 2 \cos 2\theta) \quad (2)$$

where r_0 is the tunnel radius when $e_{xx} = 0$, and θ is an angle to the horizontal (Fig. 1a).

The tilting at the tunnel wall about the y axis is given by

$$\begin{aligned} w_y &= \frac{1}{r_0} \frac{\partial r}{\partial \theta} \\ &= -4e_{xx} \sin 2\theta \end{aligned} \quad (3)$$

At the positions marked in Fig. 1b strain-coupled tilts of $\pm 4e_{xx}$ occur. Thus it is possible for strain-coupled tilts with magnitudes comparable to the theoretical body and load tidal tilts to occur. For example a linear strain tide of 1 part in 10^8 could give rise to local tilting of 4×10^{-8} rad.

Thus the value of $S_T(f)$ generated by this mechanism can take on the necessary magnitude and phase to explain the differences between the observed and theoretical values of $H_T(f)$.

It is important to position tiltmeters so as to minimize the effects of site generated tilts. The alternative approach, that of correcting the observed tilts by calculating the magnitude and phase of the strain generated tilt has many practical difficulties, primarily the difficulty of knowing the exact geometry and rigidity distribution of the cavity walls. This approach is therefore best ignored.

Horizontal pendulum type tiltmeters are often installed in niches cut into the side of underground cavities. Clearly in the ideal case (Fig. 1c) tiltmeters could be installed at positions A, B, C and D. But in real underground sites even when these are of simple geometrical form, uncertainties caused by fissures in the surrounding rock make it unrealistic to determine the exact positions of minimum strain-tilt coupling.

Provided that the tunnel is horizontal and beneath a uniform ground surface, however, tilts along the axis of the tunnel will be unaffected by tidal rock stresses. Thus when tiltmeters are sited in tunnels they should be arranged to measure tilt only along the tunnel axis. Separate tunnels should be used for

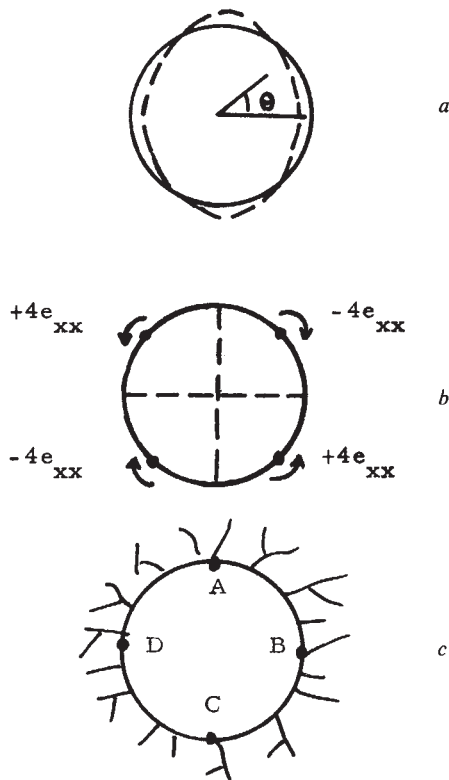


Fig. 1 Strain-tilt coupling caused by a tunnel. The y direction is along the tunnel.

the other components. Because of irregularities in the tunnel surface a suitable installation method might be to place the tiltmeter on a long beam on the floor of the cavity supported perhaps by sand at each end.

These comments apply only to pendulum tiltmeters sited in tunnels. They do not apply to borehole tiltmeters when actually used in a borehole, nor do they apply to water tube tiltmeters which are almost always mounted along the length of tunnels because of the length of water tube required to get adequate sensitivity.

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Tidal Tilt Anomalies

King and Bilham¹ have made an interesting suggestion for a mechanism to explain the large number of published anomalous tidal tilt results. The proposed coupling between the tidal strain and the tidal tilt leads to tilt anomalies of the order of 8 msec for the M_2 body tide and, for the loading tide, anomalies depending on the magnitude of the loading strain.

A mine or tunnel, in which tilt measurements are usually taken, represents a discontinuity in the Earth's crust². The strain-tilt coupling clearly provides a possible mechanism for understanding the station cavity problem. Here we present some experimental evidence which generally agrees with the predictions of the mechanism and discuss the implications.

An intensive programme of tilt measurements has been performed in the past few years, in the vault at Bidston, chiefly as a test of instrumental performance utilizing the very large loading signal (M_2 amplitude: 70 msec north-south; 55 msec east-west) before progressing to the geophysical station at Llanrwst. A variety of different designs of tiltmeter have been used: two Verbaandert/Melchior horizontal pendulums; two Tomaschek-Schaffernicht pendulums; one smaller Tomaschek pendulum; two modified Milne-Shaw pendulums; and two Askania vertical pendulums. The vault is 15 m by 2.5 m with its length precisely in the east-west azimuth. Much data has been obtained from these different instruments at sites along the length of the vault but the general conclusions are independent of the particular instrumental design. The M_2 phase is consistent in both the north-south and east-west azimuths independent of position within the vault. Similarly, the amplitude of the east-west tilt seems to be mostly independent of position within the vault even for measurements taken at the two ends of the vault³. The magnitude of the north-south tilt, however, depends on position, and varies by 20% from one end of the vault to the other.

If, for simplicity, we regard the vault as a cylindrical cavity of length 15 m and diameter 2.5 m, then the results are seen to conform to the general predictions of King and Bilham. Tilts along the axis of the cylinder are more consistent than those across the cylinder. It is interesting that the north-south variations exist even though from symmetry considerations most of the measurements are taken at the points where the anomaly may be expected to be a minimum (ref. 1: Fig. 1c, position C), showing that the real situation is indeed more complicated than these simple considerations might suggest. In fact, the vault is excavated in the sandstone below the Bidston observatory, so the local topography and general environment are perhaps too complex to justify more definitive investigations on these lines.

Work is in progress to identify such effects in the geophysical station at Llanrwst. There are already some indications from a very short time series using the prototype Askania vertical pendulum in comparison with the Tomaschek horizontal pendulums that more consistent tilts are recorded in azimuths corresponding to the axis of the tunnel.

If these indications prove to be correct then, clearly, there are many general points to be considered regarding instrument installation. Ideally, the instrument should be as far away from the wall of the cavity as is possible, at least for measuring the tilt in the azimuth normal to the wall. The common practice of using special niches cut into the rock must be viewed with caution, especially for the azimuth out of the wall. Indeed the niche itself may respond in some unknown way. Again, a vertical pendulum may be more reliably employed in measuring the tilt parallel to the wall to which it is clamped. Although the recent rapid instrumental development of tiltmeters is welcome, the most basic problem lies in the siting of the instruments rather than in the instruments themselves.

Concentration of effort in measuring the tilt along the tunnel axis may be the best policy to adopt but, in any case^{2,3}, detailed measurements of the coherency of the tidal signal over a single station should be made before attempting to discuss the regional tilt signal. In a real situation any coupling effects will be extremely complicated and can only be investigated by such a detailed experimental programme. It is worth noting that if the effects are non-linear then indication of this may be obtained by examining the higher frequency components such as M_4 and MS_4 . The use of special shafts or boreholes, although having their own problems, may be less affected by this coupling problem and may eventually provide an alternative solution.

King and Bilham¹ suggest that the strain-tilt coupling at a particular site is too difficult to calculate and that there-

fore this approach should be ignored. Nevertheless, there is much experimental information which should be reassessed in the light of this coupling, if station configuration could be ascertained. It would be interesting to consider under what circumstances the perturbation leads to a decrease of the recorded tilt as opposed to an amplification. This will presumably depend on the location of the instrument with respect to the walls of the cavity. For example, a north-south tiltmeter on a north wall will be perturbed in an opposite sense to a north-south tiltmeter on a south wall. In the Ardennes, at four different stations, anomalously low M_2 north-south tilts have been observed. Is this related to tilt/strain coupling or is there a larger scale perturbation, for example arising from the tidal effects in an aquifer as suggested by Bower⁴? One particular point of interest (G. C. P. King and R. G. Bilham, private communication) is that the mechanism may provide an explanation of the greater magnitude of the north-south M_2 diminishing factor anomalies by comparison with east-west anomalies in Europe^{2,3}.

In this area the amplitude of north-south tilt is about half that of east-west tilt, whereas theory suggests the opposite to be the case for strain. This, combined with the differing phase relationship between tilt and strain in the two directional components, would seem to provide a clue as to the explanation of the bias in the observed anomalies in Europe.

King and Bilham¹ state that amplitude anomalies are often attributed to poor instrument calibration; this is not usually possible because even poor instrument calibration rarely leads to an error greater than 8% and most of the observed anomalies are significantly larger than this. Also, they conclude that phase anomalies cannot be attributed to instrumental causes. This is an oversimplification of the case. Even with gravity meters phase errors are well known. With tiltmeters phase errors can arise from the following causes⁴: ill defined true recording azimuth of a tiltmeter; errors in surveying procedures; a drifting recording azimuth (in the case of a horizontal pendulum); and time errors. These errors are often accorded insufficient attention. The general picture of phase anomalies is probably confused by the practice of transforming from the actual recording azimuth to north-south and east-west azimuths in publishing results, so that the phase anomaly becomes a function of the amplitude anomaly and the angle of rotation of the reference azimuths.

As regards meteorological or other variable influences on tilt, the best approach is to analyse a long time series (preferably greater than 1 yr) in separate monthly sections, allowing for constituent separation problems. Only in this way can the reliability of the diurnal constituents O_1 and K_1 , and indeed other constituents, be assessed accurately for any given site.

We conclude that it now seems worthwhile to examine the present experimental evidence, in the light of the proposed mechanism, to see if a consistent pattern emerges. From the experience so gained and from further detailed experimental investigation consistent and geophysically useful tidal tilt information should ensue.

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Geochemical and Geophysical Implications of the Reaction between Seawater and the Oceanic Crust

I have presented a detailed synthesis of the evidence for large scale chemical reactions between seawater and the upper 2–3 km of the ocean floor basalt¹. These reactions seem reasonable as the ocean floor is free from thick sediment for about 80×10^6 yr and is cross-cut with fractures and joints that permit seawater to circulate freely and react with basalt over long periods of time. Here, I emphasize some of the implications of these reactions.

The evidence for the reactions is both geochemical and geophysical. The chemistry of basalts sampled on the ocean floor varies progressively with distance from principal sea-floor spreading ridges. Further, the seismic velocities of the uppermost rock layer of oceanic crust decrease progressively from the ridge, probably because of leaching of elements and hydration of the basalt layer by seawater. Similarly, the decrease in natural remnant magnetism of the seafloor with distance from the ridge can best be explained by a two layer model with the upper layer (0.5 km thick) exposed to seawater oxidation of the ferromagnetic minerals².

As a working hypothesis the rate of decrease of seismic velocity with distance from the oceanic ridges can be taken as a measure of the integrated rate of chemical weathering. Dividing this rate by the change of laboratory measured seismic velocity in basalt samples as a function of chemical changes, the rates of exchange for the principal elements can be computed. In general, the rates using the seismic velocity model are 1/3 to 1/2 the values observed by sampling basalts directly on the floor of the ocean (Fig. 1). This is expected if the rate of the basalt seawater reactions diminishes with depth.

As well as this weathering reaction, there is convincing isotopic evidence that greenschist facies metamorphic rocks have reacted with seawater, probably at greater depths than the weathering reaction^{3,4}. Fig. 2 shows the chemistry of greenstones related to fresh basalts as a function of water content. There seem to be two distinct processes. The first, a primary stage which forms under the ridge crest, is characterized by the take-up of sodium and the loss of iron; a retrograde stage is dominated by a pickup of magnesium, iron and water.

Combining the weathering reaction with the primary and

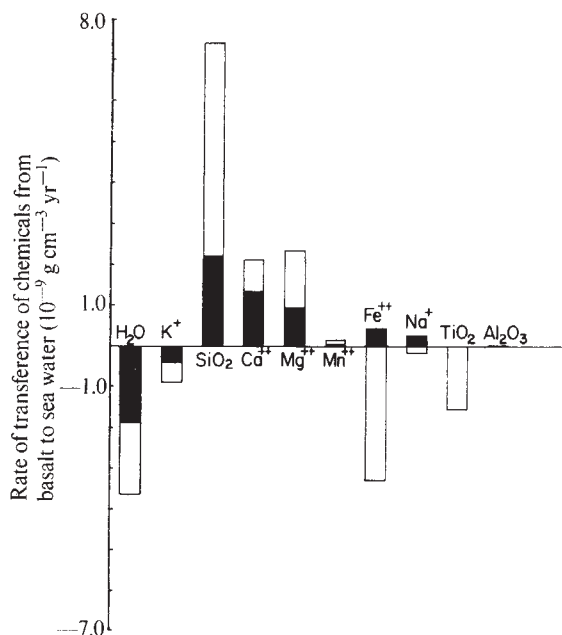


Fig. 1 □, Rate of transference calculated from surface samples; ■, rate from layer 2 seismic velocity. From ref. 1.

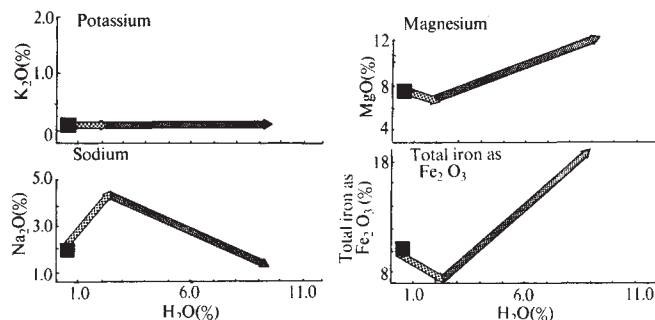
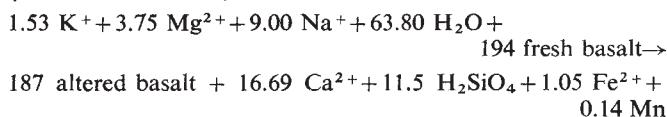


Fig. 2 Chemical changes produced by greenschist metamorphism of tholiitic ridge basalt¹. ■, Fresh ridge basalt; dotted arrow, primary greenschist metamorphism (albite-actinolite dominant); cross-hatched arrow, retrograde greenschist metamorphism (chlorite dominant).

retrograde greenschist reaction, it is possible to write a general equation for the overall reaction between seawater and basalts of the seafloor for a reaction extending to a depth of 2.5 km. (Units are 10^{12} mol yr^{-1} .)



An important implication of the balance is that about 80% of the K^+ , 60% of the Mg^{2+} and all the Na^+ from stream supply could be absorbed by the seafloor basalts while Ca^{2+} , SiO_2 , Fe^{2+} and Mn^{2+} could be released to seawater in amounts comparable to stream supply. The Ca^{2+} and SiO_2 eventually find their way into the sediment pile by the action of diatoms, while the Fe^{2+} and Mn^{2+} are probably oxidized in metal-rich sediments and in manganese nodules.

I also emphasize that this reaction is most likely exothermic, and that the energy released at standard conditions can be estimated using the standard heat of formation of the cation species in aqueous solutions.

A minimum standard heat of reaction can be obtained by assuming that the heats of formation of solid products are equal to the solid reactants and a maximum by taking products to be on the average $100 \text{ kcal mol}^{-1}$ less than the reactants. The minimum is $0.70 \times 10^{16} \text{ kcal yr}^{-1}$ and the maximum is $7.0 \times 10^{16} \text{ kcal yr}^{-1}$. It is possible to convert these values into standard heat flow units and compare them with a heat flow profile over the East Pacific Rise (Fig. 3). The observation that heat flow over the ocean floor is comparable with that measured over the more radiogenic continents

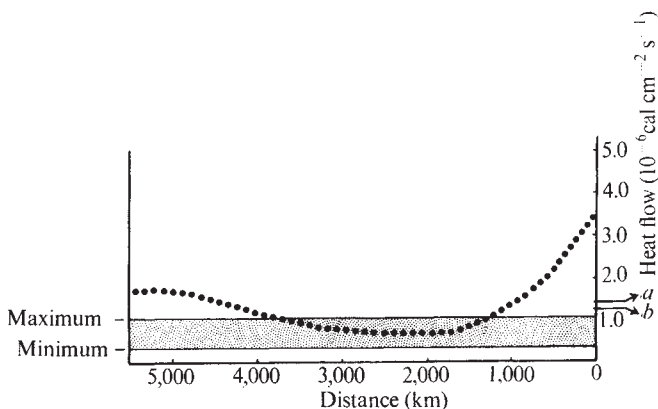


Fig. 3, Heat flow profile over East Pacific rise⁷; shaded area, limiting band of chemical energy generation regime¹. a, Pacific basin average; b, continental US average.

is consistent with the suggestion that chemical reactions on the seafloor release energy comparable with the measured heat flow.

Finally I shall consider the possibility that changes in spreading could alter the chemical balance of the ocean and atmosphere. If the rate of spreading of the floors of the oceans increased, the volume of fresh basalt in contact with seawater would be increased and the potential for the chemical reactions would also become larger. The validity of the whole concept of chemical reactions on the seafloor could be tested by studying fluctuations in the chemistry of seawater as a function of time. Unfortunately, there are very few indicators of the chemical history of water that are not affected by secondary processes such as diagenesis. Two isotopic indicators that may be useful are the $^{87}\text{Sr}/^{86}\text{Sr}$ ratios measured in marine fossils, and $^{34}\text{S}/^{32}\text{S}$ ratios measured in evaporites. The measured values for these two isotope ratios through time are shown in Fig. 4. The isotope ratios do fluctuate with time, and the fluctuations for each element may be synchronous.

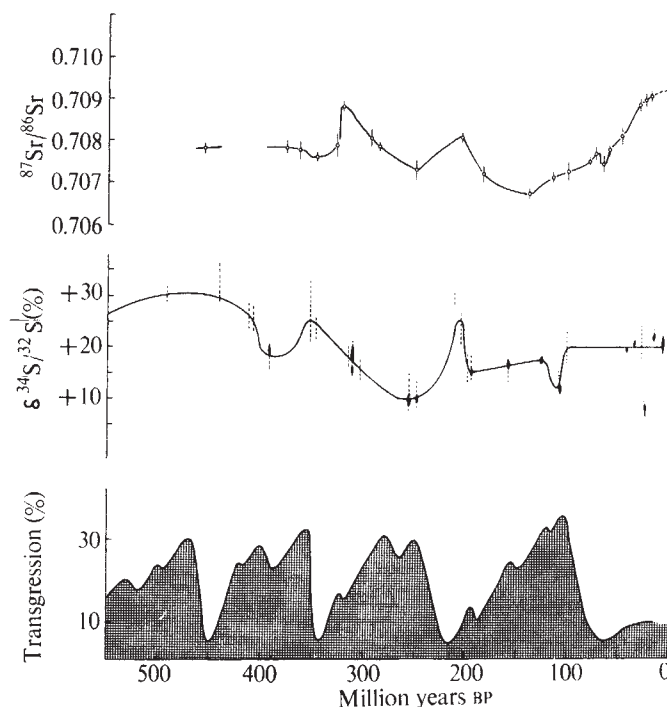


Fig. 4 Fluctuations of the strontium and sulphur compositions of seawater compared to epicontinental marine transgressions. Strontium isotope curve from ref. 8; sulphur isotope curve from Holser and Kaplan⁹; marine transgression curve from Damon¹⁰.

Is it possible that these isotope fluctuations in seawater are caused by changes in seafloor spreading? There are no direct measurements of global changes in seafloor spreading; it has, however, been suggested⁵ that times of high epicontinental sea transgression are times of high spreading rates; these are also plotted in Fig. 4.

Studies^{5,6} show that fresh ridge basalt has a lower $^{87}\text{Sr}/^{86}\text{Sr}$ ratio than basalt that has been altered by seawater. The $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of fresh ridge basalt is about 0.7026 and altered basalts have been measured with ratios as high as 0.7040. The times of fast spreading, as suggested by high marine transgression, roughly correspond to minima on the $^{87}\text{Sr}/^{86}\text{Sr}$ curve, suggesting the addition of non-radiogenic material from the basalt. Periods of slow spreading correspond, within the limitations of fossil dating, to maxima in the isotope curve because stream supply of radiogenic continental strontium overrides the supply from the basalt.

There is a similar approximate correspondence for the sulphur isotope curve, with minima on the curve matching times of faster spreading and maxima matching times of slower spreading. The explanation could be similar to the strontium case. The sulphur isotope ratios of basalt sulphide are lower than stream-supplied sulphate.

It also seems likely that changes in spreading rate would alter the residence times of K^+ , Na^+ and Mg^{2+} in seawater and alter the supply of SiO_2 , Ca^{2+} , Fe^{2+} and Mn^{2+} to seawater. In turn it is to be expected that the rate of deposition of these elements in seafloor sediments would vary. The anion species with which these elements combine during sedimentation, chiefly CO_3^{2-} and O_2 would also vary. Therefore the CO_2 and O_2 content of the atmosphere could be dependent on conditions of seafloor spreading.

There is a need for more information on global changes in spreading rate and fluctuations in the chemistry of seawater; the data now available is, at least, consistent with the hypothesis of large scale reactions between seawater and basalt on the ocean floor.

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Compressive Stress between Two Mid-Ocean Ridges

ONE of the possible driving mechanisms of plate motions¹⁻³ is a push from mid-ocean ridges. This could arise from the injection of magma at the ridge crest⁴, or to hydrostatic overpressure resulting from the elevation of the ridge above the ocean basins⁵. If the plates are pushed apart at ridges, an area between two mid-ocean ridges should be under

compression. On May 9, 1971, an earthquake of magnitude 6.2 (m_b reported by NOAA) occurred within the Antarctic plate between the Chile Ridge and the Pacific-Antarctic Ridge, followed within 24 h by several aftershocks of magnitude 5.0 or greater (Fig. 1). An analysis of the focal mechanism of the principal shock shows that the plate is at least locally under horizontal compressive stress, with the maximum stress axis approximately perpendicular to the Pacific-Antarctic Ridge.

The S-wave polarizations and the P-wave compressional arrivals (Fig. 2) are consistent with predominantly thrust faulting. But the body wave data provide little control on the orientation of the fault and auxiliary planes. According to the P-wave data, the more steeply dipping plane, which for convenience I call the fault plane, could be assigned an orientation ranging from a strike of 175° east of north and a dip of 56° to a strike of 218° and a dip of 68° . There is almost no control on the other plane. Additional constraints on the source geometry were obtained from observations of Rayleigh wave amplitudes.

The amplitude of the Rayleigh waves (seismic surface waves) depends on the strike and dip of the fault plane and the direction of the slip on the fault. A plot of amplitude as a function for azimuth for a dip-slip source yields a two-lobed radiation pattern (Fig. 2). A strike-slip event would have a four-lobed pattern. By fitting theoretical radiation patterns to the observed azimuthal dependence of Rayleigh wave amplitude, the focal mechanism can be determined.

I measured the amplitudes by Fourier analysis of the long period vertical component of seismograph records from twenty-eight WWSSN stations, digitized at intervals of about 1.0 s. The observed amplitudes, corrected for attenuation, instrument response, and geometrical spreading on a spherical Earth, are plotted in Fig. 2. The overall pattern is two-lobed, reflecting the dip-slip nature of the event. Theoretical radiation patterns were generated according to formulas given by Saito⁶ for the excitation of surface waves in a layered Earth, using an oceanic Earth model⁷. The least squares fit of the theoretical pattern to the observed amplitudes was computed for each source model. With the dip of the fault plane of approximately 60° determined by the body wave data, the slip and strike were allowed to vary until a number of acceptable models were found. A statistical *F*-test was performed comparing the fit of the best model with the fit of all other models. If, according to the *F*-test, there was a 90% probability that the best model was significantly better than some other model, then the other model was rejected. In this way, a range of acceptable models was outlined, with the boundaries defined at the 90% confidence limits. The region of possible models of the principal shock is shown in Fig. 3, with the 95% confidence limits also outlined.

Although the Rayleigh wave data do not yield any

Fig. 1 Location of the earthquakes of May 9, 1971. Epicentre of the principal shock is given by NOAA earthquake data report as 39.78° S, 104.84° W. The arrows indicate the direction of maximum horizontal compressive stress. Double lines represent actively spreading ridges, single lines are transform faults. The details of the plate boundaries in the Chile Fracture Zone are not well known. The strike of the Chile Ridge may be more north-south than is shown here (K. Klitgord, personal communication).

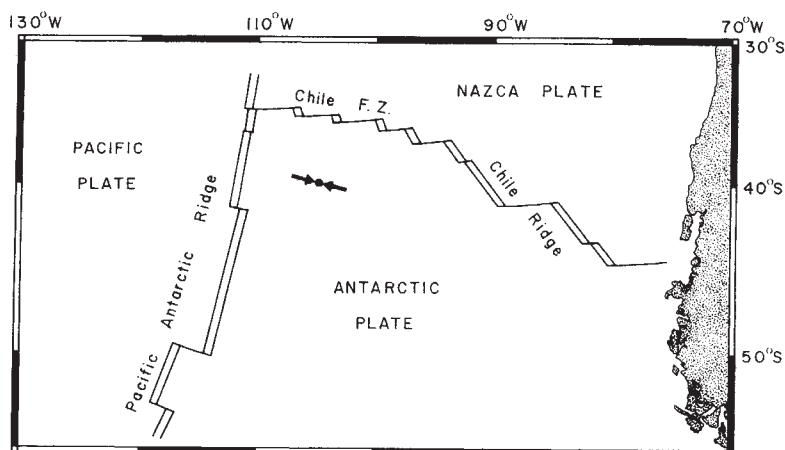
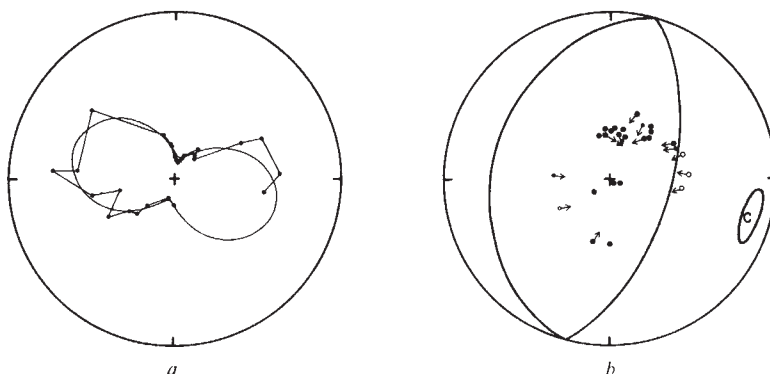


Fig. 2 Focal mechanism of the principal shock. In *a*, dots indicate observed amplitudes of 67 s Rayleigh waves as function of azimuth. The amplitude is proportional to the distance from the centre of the figure. The smooth continuous line shows the theoretical radiation pattern. *b* is a Schmidt net projection of the lower focal hemisphere showing the distribution of P-wave polarities and the direction of polarization of the S-waves. ●, Compressions; ○, dilatations. Smaller symbols represent poorer data. The small elliptical area surrounding the "C" indicates the uncertainty in the compressional axis. $Q=125$; strike= 196° ; dip= 60° ; slip= 90° , depth=5.



additional limitation on the strike, the slip is found to be limited to $90 \pm 25^\circ$. A slip of 90° represents pure thrusting, and a slip of 0° or 180° is pure strike-slip. But there is now the added constraint that, for any given strike, the uncertainty in slip is only about $\pm 10^\circ$. This constraint greatly reduces the uncertainty in the direction of the compression axis. The uncertainty in the compression axis is indicated by the ellipse in Fig. 2*b*. The strike of the compression axis is $105 \pm 11^\circ$ east of north. The focal mechanism shown in Fig. 2 is the solution closest to the centre of the region of uncertainty.

In theory, the depth of the earthquake can be determined from the change in the radiation pattern with decreasing period of the Rayleigh wave. Because there is a large degree of scatter in the amplitudes for periods less than 20 s, the depth could not be precisely determined. Although the data seem to indicate that the shock was very shallow, less than 10 km, a detailed study of the effects of multiple path interference and attenuation would be required to determine the depth with certainty.

The event of May 9, 1971, is ideally located for a test of the hypothesis that plates are pushed apart at mid-ocean ridges. It is in perhaps the only area between two roughly parallel, actively spreading ridges, effectively isolating it from all other global tectonic processes. It is difficult to imagine the stress being generated by two convection cells upwelling

under the ridge crests and flowing toward each other, without some surface expression at the point of juncture of the cells. The stress cannot be transmitted from another oceanic plate, because the ridge crests are tensile features decoupling the plates. The strike of the Pacific-Antarctic Ridge at 40° S is about 10° north of east⁸, within 5° of being perpendicular to the axis of compressive stress. This observation and that of Mendiguren⁹ of compressive stress within the Nazca plate can be explained most simply by failure at a weak point of a lithosphere under horizontal compressive stress caused by pushing from the mid-ocean ridges. A geographical traverse, which passed very close to the epicentre, found no obvious local tectonic feature which could create a local stress field¹⁰. Thus, the stress field found supports the hypothesis that pushing from the ridges is a primary driving mechanism of plate tectonics.

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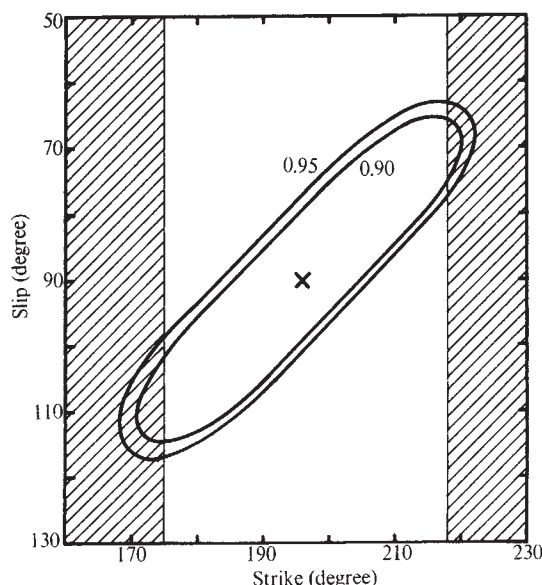
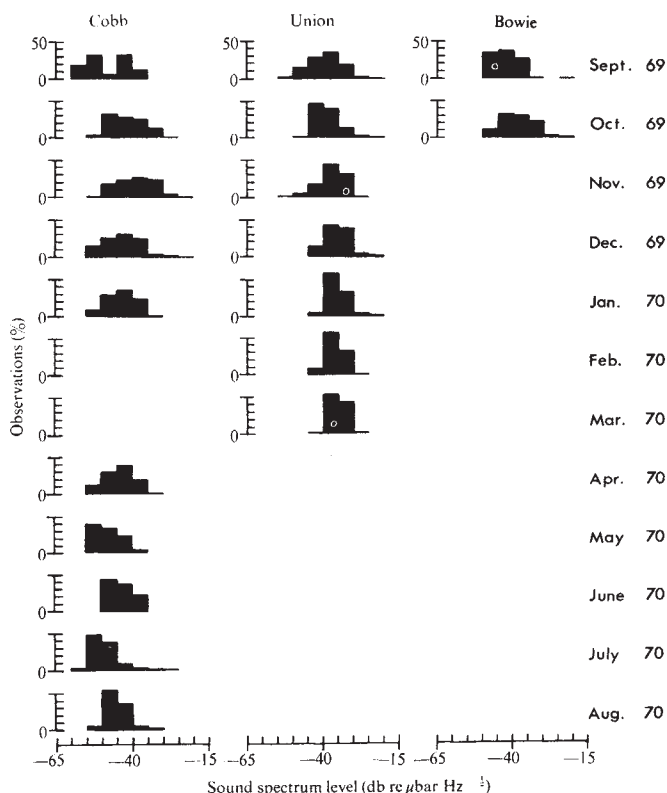
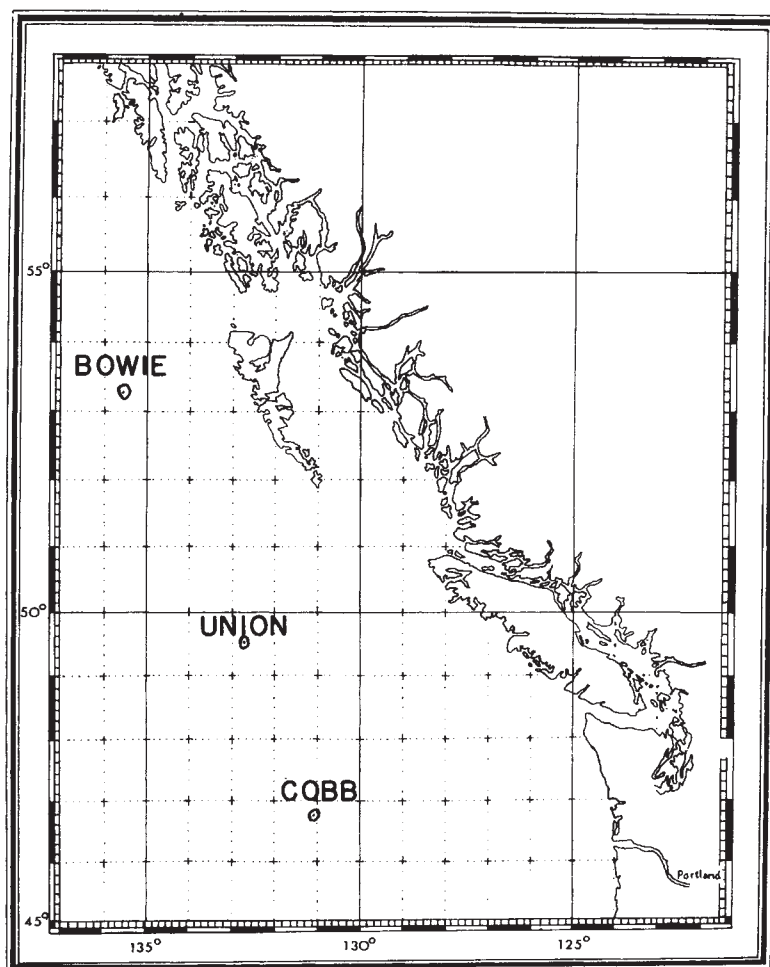


Fig. 3 Region of uncertainty in the fault plane solution. Based on analysis of 67 s period Rayleigh waves, only models with strike and slip within the inner elliptical region are considered acceptable. Models within the shaded regions are eliminated by the body wave data. The slip angle is measured counterclockwise from a horizontal line on the fault plane pointing in the direction of the strike. ×, Centre of region of uncertainty. Dip is 60° .

Seasonal Variation of Underwater Noise in North-east Pacific

BEFORE marine seismic and other systems that rely on the detection of acoustic signals can be successfully designed, variation of ambient acoustic levels in the oceans throughout the year must be determined. Here I report measurements of ambient noise over a 1 yr period made recently at three sites in the North-east Pacific (Fig. 1). Remote instrument packages designed for a maximum water depth of 670 m, as previously used in relatively shallow Arctic waters¹, were placed near the summits of seamounts Bowie (summit depth about 40 m)², Union (~ 320 m)² and Cobb (~ 34 m)³. The package on

Fig. 1 Chart of the North-east Pacific showing the location of Seamounts Bowie, Union and Cobb.



Bowie was placed in a saddle—at the north-east end of the approximately 4 mile long ridge-like summit at a depth of 220 m. On Union the package was placed within 0.5 mile of a dome-like summit at a depth of 340 m and on Cobb the package was placed on a gently sloping terrace some 2 mile to the east of the summit, at a depth of 224 m. Acoustic measurements were limited to the frequency band 150 to 300 Hz. The data yield, although limited by equipment malfunction, was sufficient to reveal seasonal variations.

Four-minute observations of the noise sound pressure level were made every hour, averaged approximately using analogue circuitry over the observation period, and recorded as a single digital sample. Sampled values are presented in Fig. 2. An increase in noise level, which can be attributed at these acoustic frequencies almost entirely to storm activity and the associated high wind speeds, occurs in the winter months, although this trend is largely masked by the 3 to 6 dB standard deviation in the format of Fig. 2. The seasonal variation is best illustrated by plotting the monthly mean noise levels over the one-year observation period (Fig. 3). As well as the obvious seasonal trends, a uniform shift of about 8 dB is seen in the noise levels observed at Union Seamount relative to those observed at Cobb. This may well be attributable to the higher wind speeds likely to be encountered at the more northerly latitude of Union Seamount. But the limited data available from Bowie Seamount do not bear out this trend; the levels recorded at this site, the most northerly, fall between those measured at

Fig. 2 Histograms of frequency of occurrence of ambient noise level for 5 dB increments of level for each month of the year and for each seamount. Equipment failure limited the useful data obtained at Union and Bowie Seamounts, and also for February and March, at Cobb.

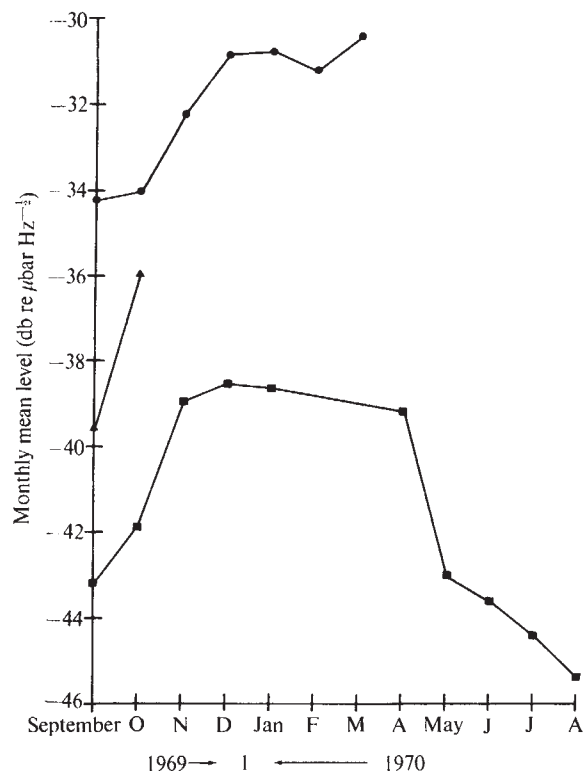


Fig. 3 Monthly mean ambient noise levels from September 1969 to August 1970. ●, Union; ▲, Bowie; ■, Cobb.

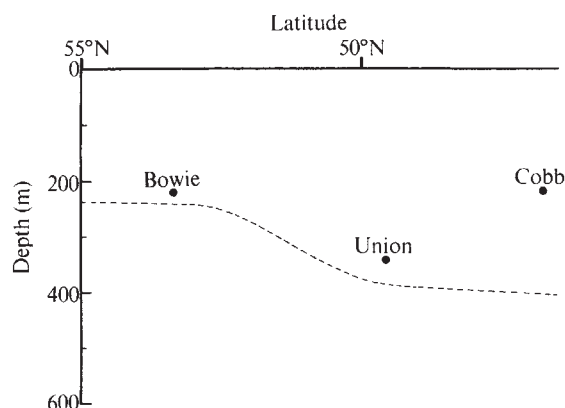


Fig. 4 Deep sound channel axis depth against latitude along an approximately straight line through the positions of seamounts Bowie, Union and Cobb. Instrument package latitudes and depth for the three seamounts are also plotted.

Union and Cobb Seamounts. The difference in prevailing noise levels may also be attributable to the different depths at which the sensors were located on the two seamounts and in particular to the fact that the deeper unit, located on Union, was situated close to the axis of the deep sound channel (Fig. 4). Under these circumstances an enhancement of ambient noise level at the depth of the sensor placed on Union Seamount is consistent with a hypothesis that much of the energy associated with underwater ambient noise is trapped in the sound channel.

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Point Defects and Extended Defects in Niobium Oxides

THE part played by true point defects—isolated vacancies and interstitial atoms—in defect solid solutions and in mixed valence non-stoichiometric phases is not clear. In many ionic or quasi-ionic structures there is some structural reorganization and at least incipient ordering of defect clusters. In some oxides—notably those of titanium, vanadium, molybdenum and tungsten, in which the covalent component of the bonding is probably strong—slight reduction, with a change in the ratio of oxygen to metal, is accommodated largely by crystallographic shear. A complete sheet of anion sites is eliminated from the structure, forming an extended defect and greatly diminishing the extent to which a deficit of oxygen must be compensated by anion vacancies or interstitial cations. Thus Hyde and Bursill¹ found that, in slightly reduced rutile, $\text{TiO}_{1.9986}$, the point defect concentration could hardly exceed 10^{-4} . In more extensively reduced rutile, changes in stoichiometry involve only changes in the spacing between crystallographic shear planes, without detectable concentrations of localized defects.

How far is the same true of the tightly defined crystal chemistry of the so-called "block structure" oxides, typified by niobium pentoxide? These constitute an extensive family of binary and ternary oxides, with structures essentially of the ReO_3 type, in which the anion/cation ratio is adjusted by crystallographic shear on (100) and (001) of the ReO_3 substructure. The whole is thereby divided into rectangular columns of ReO_3 structure, ($m \times n$) coordination octahedra in cross-section, interfaced by crystallographic shear planes² (Fig. 1); these columns constitute subunits or building blocks that can be assembled into a wide variety of ordered or disordered structures with corresponding gradations of total composition. Much evidence is now available from lattice imaging electron microscopy to show this³⁻⁷. The dimensions ($m \times n$) of the columns, and the way columns are linked at their corners, completely specify the stoichiometry of any perfect structure, but because it is topologically possible for different block structures, with different compositions, to intergrow coherently, very subtle changes of stoichiometry can be accommodated by the occasional intergrowth of lamellae of different structure. Fig. 2 shows one such extended defect: a lamella of $\text{H-Nb}_2\text{O}_5$ structure in a matrix of $\text{MgNb}_{24}\text{O}_{60}\text{F}_2$ (isostructural with $\text{Nb}_{25}\text{O}_{62}$). If no other, more localized, defects can be formed, it should be possible to account for deviations from the ideal composition of a given structure entirely in terms of coherent intergrowths.

It is, therefore, important that there is considerable evidence for variability of composition, in certain of these phases, that cannot be so explained. Gruehn⁸ has shown that block structures with tetrahedrally coordinated cations in tunnels—and only those compounds—can have an analytically measurable, monophasic range, which does not necessarily include the ideal composition (Table 1). In a systematic lattice imaging study of the $\text{WO}_3\text{-Nb}_2\text{O}_5$ system, Allpress and Roth⁹ could find no evidence of coherently intergrown "faults" in the $(4 \times 3)_1$

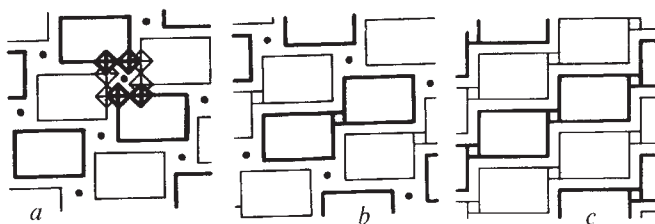


Fig. 1 a, Idealized structure of $\text{WNb}_{12}\text{O}_{33}$, [010] projection showing columns of octahedra, (4×3) in cross-section and the detailed structure of the tetrahedral site (●); b, structure of $\text{Nb}_{25}\text{O}_{62}$, [010] projection; c, structure of $\text{Nb}_{12}\text{O}_{29}$, [010] projection.

Table 1 Occupation of Tunnel Sites in Non-stoichiometric Block Structures

	Ideal	Ratio of O to M Observed limits	Composition for perfect column structure	Maximum occupancy number for tunnel sites
$\text{W}\text{Nb}_{12}\text{O}_{33}$	2.5385	2.529–2.538	$M_{13.05}\text{O}_{33}$	1.05
$\text{Nb}_{28}\text{O}_{70}$	2.5000	2.490 or 2.495–2.500	$M_{28.12}\text{O}_{70}$ or $M_{28.06}\text{O}_{70}$	1.12–1.06
$\left. \begin{array}{l} \text{TiNb}_{52}\text{O}_{132} \\ \text{Nb}_{53}\text{O}_{132} \end{array} \right\}$	2.4906	2.483	$M_{53.16}\text{O}_{132}$	1.08
$\left. \begin{array}{l} \text{TiNb}_{24}\text{O}_{62} \\ \text{Nb}_{25}\text{O}_{62} \end{array} \right\}$	2.4800	2.472–2.478	$M_{25.08}\text{O}_{62}$ to $M_{25.02}\text{O}_{62}$	1.08 to 1.02
$\text{Nb}_{47}\text{O}_{116}$	2.4681	2.460 2.464–2.467	$M_{47.15}\text{O}_{116}$ $M_{47.08}\text{O}_{116}$ to $M_{47.02}\text{O}_{116}$	1.075 1.04 to 1.01
$\text{Nb}_{22}\text{O}_{54}$	2.4545	2.453	$M_{22.014}\text{O}_{54}$	1.01
" $\text{GeO}_2.9\text{Nb}_2\text{O}_5$ "	2.5000	2.474	$M_{10.11}\text{O}_{25}$	1.11
" $\text{P}_2\text{O}_5.22\text{Nb}_2\text{O}_5$ "	2.521	2.5000 (ref. 14)	$M_{23.20}\text{O}_{58}$	1.06

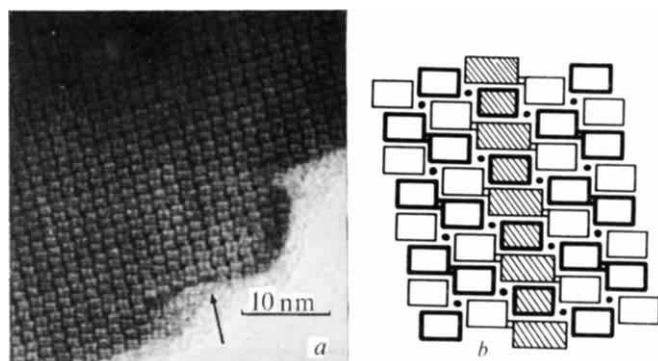


Fig. 2 *a*, Lattice image of $\text{MgNb}_{24}\text{O}_{60}\text{F}_2$ showing a lamella of $\text{H-Nb}_2\text{O}_5$ (arrowed); *b*, idealized structure of defect in $\text{MgNb}_{24}\text{O}_{60}\text{F}_2$.

block structure of $\text{W}\text{Nb}_{12}\text{O}_{33}$ over the composition range $\text{WO}_3.6\text{Nb}_2\text{O}_5$ to $\text{WO}_3.8\text{Nb}_2\text{O}_5$ ($\equiv M_{13.05}\text{O}_{33}$). In this laboratory we have found no intergrowths of $\text{Nb}_{25}\text{O}_{62}$ structure ($\text{NbO}_{2.480}$) in slightly substoichiometric $\text{H-Nb}_2\text{O}_5$, down at least to the composition $\text{NbO}_{2.495}$. All of these cases thus appear to represent genuine cation-excess structures.

An unambiguous example of an inherently defective block-structure is furnished by the phase " $\text{GeO}_2.9\text{Nb}_2\text{O}_5$ " which Levin¹⁰, from X-ray powder diffraction data, described as being isostructural with the $M_2\text{O}_5.9\text{Nb}_2\text{O}_5$ phases ($M = \text{P, As, V, Nb}$ (ref. 11)). From the crystal structure of $\text{PNb}_9\text{O}_{25}$ (ref. 12), these are $(3 \times 3)_1$ block structures (Fig. 3), with the atoms M occupying all the tetrahedral sites. As extreme possibilities, if the cation lattice in the germanium compound were complete, the formula would be $M_{10.24}\text{O}_{74}$; if the oxygen lattice were filled, $M_{10.11}\text{O}_{25}$. There must be either oxygen vacancies, or metal "interstitials," or both. We have, therefore, made a thorough study of this compound by lattice imaging electron microscopy, neutron diffraction and X-ray diffraction methods. Full results will be published elsewhere but some conclusions bearing on stoichiometric variability in block structure oxides, and on tunnel structures in general, may be reported now.

" $\text{GeNb}_{18}\text{O}_{47}$ " is indeed statistically isostructural with $\text{PNb}_9\text{O}_{25}$, and its defect composition is not the consequence of coherent intergrowth with some other block structure. The (3×3) columns of ReO_3 structure are essentially perfect; oxygen sites and cation sites are fully occupied, and all the cations in the column are niobium. Hence the irrational composition is entirely due to an excess of cations within the tunnels. Examination of the structure shows that, instead of tetrahedral sites at $z = 1/4$ and $3/4$, cations could occupy octahedral sites at $z = 0$ or $1/2$; such sites occur in pairs, in two orientations. Our structure determinations prove that these octahedral tunnel sites are partially occupied. Cations in

these positions, bridging the gap between columns at the same level (Fig. 4), must exclude the occupation of adjacent tetrahedral sites, and the limiting composition (determined from the occupation numbers) and structural considerations are compatible with (but do not prove) the hypothesis that tetrahedral cations and paired octahedral cations are ordered in regular sequence within each tunnel. There is no sign of superlattice ordering along the tunnel direction (the b axis of the structure); hence ordering within one tunnel must be quite uncorrelated with that in neighbouring tunnels.

Table 2 Cation Deficiencies on Tunnel Sites in Block Structures

Initial compound	Oxidation product	Composition if perfect block structure retained	Occupancy of tunnel sites
$\text{Nb}_{22}\text{O}_{54}$	OxIII " $\text{Nb}_{22}\text{O}_{55}$ "	$\text{Nb}_{21.6}\text{O}_{54}$	0.6
$\text{Nb}_{47}\text{O}_{116}$	OxIV " $\text{Nb}_{47}\text{O}_{117.5}$ "	$\text{Nb}_{46.4}\text{O}_{116}$	0.7
$\text{Nb}_{25}\text{O}_{62}$	OxV " $\text{Nb}_{25}\text{O}_{62.5}$ "	$\text{Nb}_{24.8}\text{O}_{62}$	0.8
$\text{Nb}_{53}\text{O}_{132}$	OxVI " $\text{Nb}_{53}\text{O}_{132.5}$ "	$\text{Nb}_{52.8}\text{O}_{132}$	0.9

These findings make it worth examining the hypothesis that, in all these compounds, the ReO_3 -like columns are perfect in structure, and that the non-stoichiometry recorded in Table 1 is due, in each case, to the "stuffing" of tunnels in the same way as in " $\text{GeNb}_{18}\text{O}_{47}$ ". That stoichiometric variability is confined to the tunnel structures would thereby be explained. Column 3 (Table 1) shows the compositions calculated on the basis of a perfect oxygen sublattice; the consequential degree of maximum occupation of tunnel sites (column 4) is much the same as in the germanium compound. Having regard to some uncertainties in the real limiting compositions, this suggests that the limit is set by attainment of the same kind of ordering between octahedral and tetrahedral sites within each tunnel.

Tunnels can also be partially emptied of tetrahedral cations. Schäfer¹³ showed that the reduced block structure oxides $\text{Nb}_{12}\text{O}_{29}$, $\text{Nb}_{22}\text{O}_{54}$, $\text{Nb}_{47}\text{O}_{116}$, $\text{Nb}_{25}\text{O}_{62}$ and $\text{Nb}_{53}\text{O}_{132}$ could be oxidized, each to its own distinctive modification of Nb_2O_5 . Oxidation of $\text{Nb}_{12}\text{O}_{29}$, in which there are no tunnels of tetra-

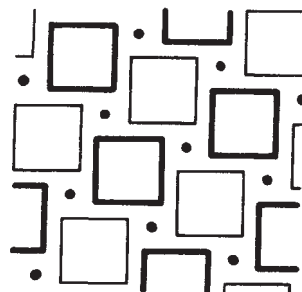


Fig. 3 Idealized structure of $\text{PNb}_9\text{O}_{25}$.

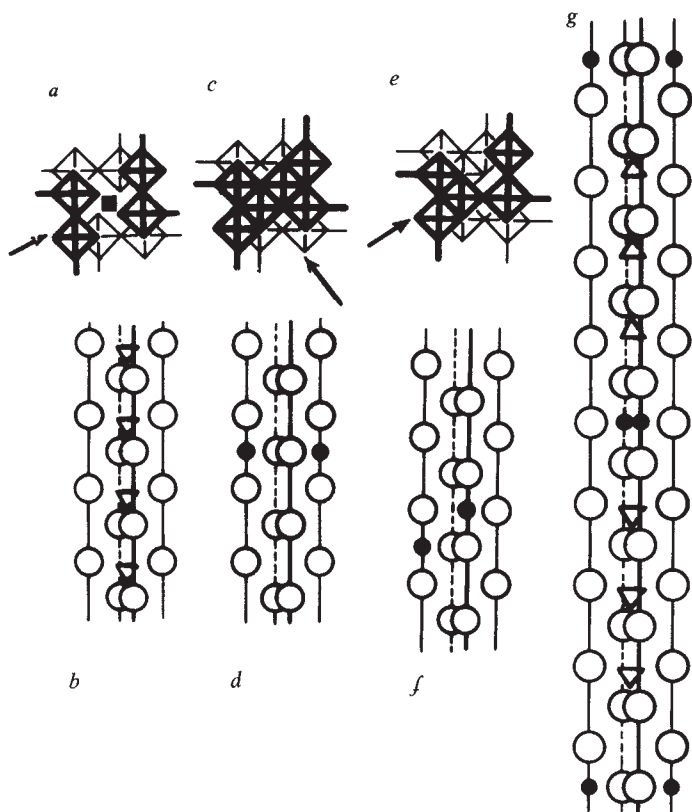


Fig. 4 *a*, Schematic projection of a tetrahedral site (■); *b*, projection of tetrahedral sites (▽) viewed in direction of arrow in (*a*). Oxygen atoms, (○); *c*, schematic projection of a pair of filled octahedral sites on one level; *d*, projection of filled octahedral sites (●) viewed in direction of arrow in (*c*); *e*, schematic projection of a pair of filled octahedral sites on different levels; *f*, projection of filled octahedral sites viewed in direction of arrow in (*e*); *g*, structure of "GeO₂.9Nb₂O₅".

hedra cations, follows a unique course¹¹. We find, from electron diffraction and lattice imaging studies that the other materials, oxidized at low temperatures to avoid subsequent partial transformation to the thermodynamically stable H-Nb₂O₅, are crystallographically identical before and after oxidation. The block structures are unaltered. On the hypothesis that the ReO₃-like columns retain their integrity, Schäfer's forms of Nb₂O₅, OxIII to OxVI, must be represented as cation-deficient, as was proposed by Wadsley and Andersson² (Table 2). Whether filled and vacant cation sites order themselves within each tunnel is uncertain. No superlattice ordering has been detected after very long annealing periods; correlation between neighbouring tunnels is clearly very small, as in the case of tunnel stuffing.

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BIOLOGICAL SCIENCES

Immunogenic Properties of Solubilized Tumour Antigen from an RNA Virus-transformed Neoplasm

NUMEROUS attempts have been made to solubilize tumour specific transplantation antigens (TSTA) of carcinogen-induced neoplasms and of virus-specified tumour antigens (VSTA) from the cell membranes of DNA and RNA virus-induced neoplasms¹⁻⁶. Usually, yields of biologically active materials are low and the materials are labile. Soluble antigenic materials obtained from transplantable hepatomas^{1,2} and purified by gel filtration were found to have limited biological activity. Although immunological specificity was preserved, the assays used were of relatively low precision. Tumour rejection, a major function of both TSTA and VSTA, was not observed.

Smith *et al.*³ claimed the "first successful solubilization and fractionation of virus-induced TSTA". Inhibition of cytotoxicity *in vitro* was reported using 'Sephadex' G150 fractions of their antigen. But this activity was distributed over a wide range of low protein concentration and high molecular weight, so that no judgment of the molecular characteristic of this material can be made.

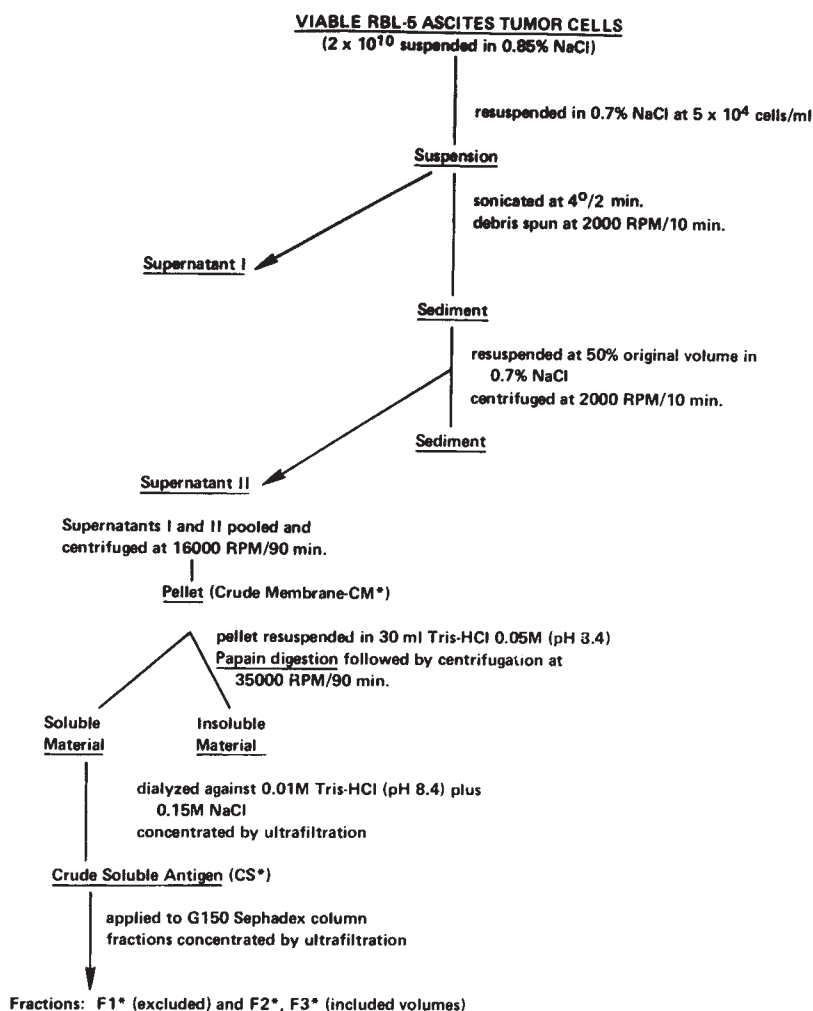
Crude extracts obtained by high speed centrifugation from tumours induced by carcinogens or by adenovirus 12 were reported to produce transplantation immunity⁴⁻⁶. It is improbable that "soluble" TSTAs were obtained by this procedure. Further purification of these materials has not been reported.

We have solubilized and partially purified histocompatibility antigens (H-2) of mice. After limited papain digestion followed by 'Sephadex' G150 column fractionation full biological activity was preserved and confined to a single peak⁷⁻⁹. These studies have provided, in our view, an excellent model and standard reference for the isolation and investigation of the immunological potentialities of soluble TSTA and VSTA.

Here we present evidence that a soluble tumour antigen isolated from cell membranes of RBL-5 leukaemic cells and partially purified can immunize animals so that specific tumour rejection occurs. The immunogenicity has been preserved through solubilization and purification.

RBL-5 leukaemic lymphocytes used as our source of tumour antigen (VSTA) have been passaged continuously in the ascitic form. This neoplasm arose in a C57Bl/6 mouse infected with Rauscher leukaemogenic virus (RLV). RBL-5 cells are immunogenic and provide immunity against other leukaemias of the FMR group (personal communication from

Fig. 1 Flow chart of solubilization and chromatography on 'Sephadex' G150 of RBL-5 VSTA from crude membranes. (Details are given in the text.) Asterisks signify the materials assayed in this study.



J. L. McCoy). RBL-5 is also a good target cell; it is easily lysed *in vitro* by sensitized syngeneic lymphocytes¹⁰, and, as determined in this laboratory, is lysed by specific cytotoxic antisera in the presence of complement; RBL-5 is not, however, very immunosensitive *in vivo* since immunized animals live only 1 or 2 d longer than controls. We have therefore used as challenge neoplasms in our *in vivo* assays LSTRA, an MLV (Moloney leukaemogenic virus)-induced leukaemia¹¹ and FBL-3, a Friend virus-induced leukaemia¹². Both are known to contain virus-specified tumour rejection antigens (VSTA) of the FMR group¹³. CBF₁ mice (Balb/c × C57Bl/6), 12–16 weeks old, were used for tumour rejection studies since LSTRA is of Balb/c origin and FBL-3 is of C57Bl/6 origin; both leukaemias grew equally well in these F₁ hosts.

The methods used for extraction and further purification of soluble VSTA from RBL-5 membranes were similar to those used and tested in our studies of H-2 antigens^{7–9} and are shown schematically in Fig. 1.

Ascites cells were collected routinely from 200 C57Bl/6 mice and red blood cells (RBC) were removed by Boyle's method¹⁴. Cells were washed twice with Tris-HCl, pH 7.4, 0.005 M; MgSO₄, 2 × 10^{−4} M; NaCl 0.85% (TBS), and their concentration was adjusted to 5 × 10⁴ cells ml^{−1} in TBS solution containing 0.7% NaCl. A sonifier cell disruptor, Model W1850 (Heat Systems-Ultra Sonics, Inc., Plainview, New York) was used to disrupt the cells, which were then centrifuged at 600g for 10 min and the sediment resuspended in TBS-0.7% NaCl at 50% of the original volume. After 1 h the suspension was centrifuged as before. Supernatants were combined and centrifuged at 55,000g for 90 min. Pellets obtained were resuspended in 0.05 M Tris buffer, pH 8.4, at approximately 14 mg ml^{−1} and digested with papain at an enzyme: crude

membrane ratio of 1 : 50 for 1 h at 37° C. The papain was activated with dithiothreitol at a final concentration of 0.01 M; the reaction was stopped with iodoacetamide at a final concentration of 0.022 M. The supernatant was separated from the insoluble sediment by centrifugation at 105,000g and was subsequently dialysed against 0.01 M Tris-HCl, pH 8.2, and 0.15 M NaCl before application to 'Sephadex' G150. The papain-solubilized material was concentrated by ultrafiltration using a UM-2 membrane (Amicon Corp., Lexington, Mass.). Protein determinations were carried out by dry weight and by the method of Lowry *et al.*¹⁵. An asterisk (*) denotes the materials in Fig. 1 that were used in our *in vivo* and *in vitro* studies.

Significant inhibition of the growth of both leukaemias, LSTRA (of Balb/c origin) and FBL-3 (of C57Bl/6 origin) in syngeneic CBF₁ mice was observed after immunization with crude membrane (CM), with the papain-solubilized CS preparation and with the F2 fraction obtained from 'Sephadex' G50 chromatography (Figs. 2–4). The F1 and F3 fractions used at the same concentrations as F2 were not immunogenic. These inhibitory effects were attained using tumour cell dosages for both leukaemias considerably above the tumour dose-50 (TD₅₀). The CM and CS preparations of RBL-5 cell membranes, administered in complete Freund's adjuvant, provided striking inhibition of the subcutaneous growths of LSTRA leukaemic cells as shown in Figs. 2A and B. The mean age to death was prolonged by 14.5 ± 2.9 d in mice treated with our CM antigen preparation (*P* < 0.001) and 11.8 ± 3.48 d with the CS antigen preparation (*P* < 0.001), comparing the group immunized with 100 µg with the Freund's adjuvant-treated controls. Inhibition of LSTRA growth by the CS preparation was also evident in the average diameter of the

growths; for example, 10 d after LSTRA challenge the average diameter of the subcutaneous growths in adjuvant controls was 5.76 mm, of CBF₁ mice immunized with 100 µg CS, 3.26 mm, and of the group immunized with 50 µg CS, 3.28 mm.

When studying the inhibitory influence of 'Sephadex' G150 fractions on the growth of LSTRA leukaemic cells, advantage was taken of the delayed rate of growth of the localized subcutaneous growth as well as the reduced frequency of takes (Fig. 3A, B and C). Cumulative units of growth as used in studies summarized in Fig. 3 represent the product of the number of progressive takes per group by mean tumour diameter with time (see legend of Fig. 3). In Fig. 3A, crude soluble antigen (CS) was again found to provide inhibition. At day 8, after leukaemic cell challenge, for example, LSTRA had reached its maximum growth in the adjuvant-treated controls whereas CS antigen-treated mice had no tumours. Resistance to the growth of LSTRA in mice pretreated with the 'Sephadex' G150 fraction, F2, is summarized in Fig. 3B and C. Those BDF₁ mice inoculated with 4.4×10^3 LSTRA cells and pretreated with the F2 fraction (Fig. 3C) showed a reduced frequency of takes and slower tumour growth when compared with controls and those mice pretreated with the F1 and F3 'Sephadex' fractions. The difference in mean units of growth observed 20 d after LSTRA challenge, between the F2 antigen recipients and the other three groups combined was 0.85 ± 0.40 ($P < 0.04$); also two out of seven mice did not grow LSTRA whereas LSTRA grew progressively and killed nineteen out of twenty-one mice of the combined groups.

FBL-3 lymphocytic leukaemia induced by Friend virus and carried as an ascitic neoplasm in C57Bl/6 mice is known to contain type-C virus particles and possesses membrane tumour antigens (cellular or virion) which cross-react with other leukaemias induced by the FMR group; this leukaemia

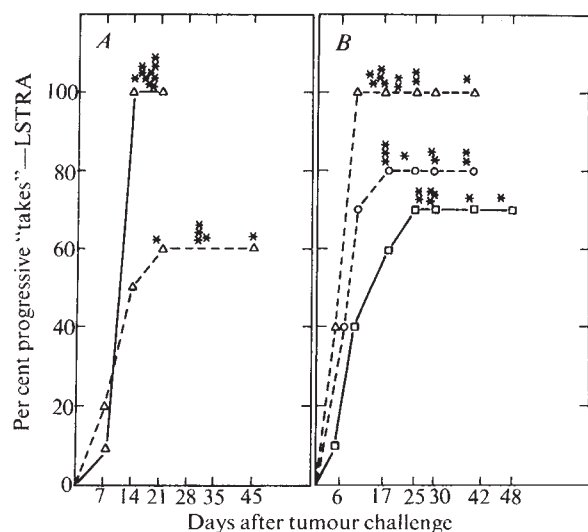


Fig. 2 Inhibition of growth of LSTRA leukaemia in 12-week-old CBF₁ female recipients. Data in A represent combined groups receiving 2.9×10^3 and 5.8×10^3 ascitic cells inoculated subcutaneously; data in B represent combined groups inoculated subcutaneously with 4.4×10^3 and 2.2×10^3 cells respectively. The TD_{50} for LSTRA leukaemic cells is $< 1.0 \times 10^3$ cells. A: Δ --- Δ , mice inoculated intraperitoneally with 400 µg CM antigen in complete Freund's adjuvant from RBL-5 leukaemic cells; two antigen inoculations were given 7 d apart and challenge with LSTRA was made 14 d after the last immunization. Control mice (Δ — Δ) received Freund's adjuvant. Asterisks above curves represent days to death with neoplasm. The mean time to death in CM-immunized mice was 32.0 ± 2.9 d and in controls, 17.5 ± 0.51 d. The period of observation was 90 d. B: $\bigcirc$ --- $\bigcirc$, CBF₁ female mice immunized with 50 µg of crude soluble (CS) antigen in Freund's adjuvant $\times 3$ at weekly intervals; $\square$ — $\square$, immunization with 100 µg CS $\times 3$; Δ --- Δ , control mice inoculated with Freund's adjuvant only. The mean time to death of the control group was 20.5 ± 2.2 d, of the 50 µg group, 26.0 d and of the 100 µg group, 32.3 ± 2.7 d.

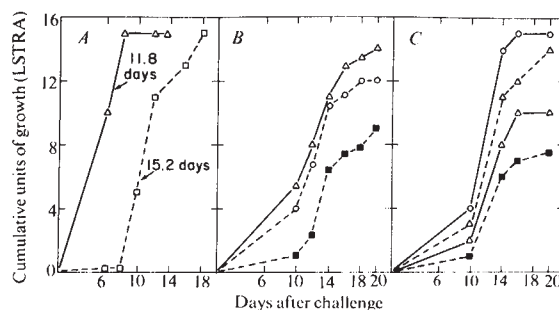


Fig. 3 Inhibition of the growth of LSTRA in CBF₁ female mice 12 weeks old immunized with antigen and with 'Sephadex' G150 fractions: F1, F2 and F3. Inhibition of growth was reflected in the rate of growth of the localized tumour as well as in the frequency of progressively growing takes; thus, cumulative units of growth represent the product of the number of takes per group by the mean average diameter of the subcutaneous growth. Units of mean diameter, used for simplification, were: 0.5 (5 $\times$ 5 $\times$ 5 mm), 1 (10 $\times$ 10 $\times$ 15 mm), 2 (15 $\times$ 15 $\times$ 25 mm) and 3 (25 $\times$ 25 $\times$ 40 mm or greater). Maximum growth obtained in five mice per group was therefore 15 cumulative units. A: $\square$ --- $\square$, five mice inoculated with 50 µg CS antigen in complete Freund's adjuvant weekly $\times 3$; Δ — Δ , five control mice received complete Freund's adjuvant only. Challenge, subcutaneously with 2.3×10^3 LSTRA cells was made 2 weeks after the last immunization. The mean time to death of immunized mice was 15.2 d and of control mice, 11.8 d. B: $\blacksquare$ --- $\blacksquare$, CBF₁ mice immunized with the 'Sephadex' G150, F2 fraction, 100 µg in complete Freund's adjuvant inoculated $\times 2$, at 1 week intervals; $\bigcirc$ --- $\bigcirc$, mice inoculated with the F3 fraction at 100 µg $\times 2$; Δ — Δ , controls received complete Freund's adjuvant. There were ten mice in each group, half of which were inoculated subcutaneously 2 weeks after the last treatment with 1.2×10^4 LSTRA leukaemic cells and half of which received 6×10^3 cells. Thus, cumulative units of growth represent the means of these two subgroups of five each within each group. C: $\blacksquare$ --- $\blacksquare$, CBF₁ mice inoculated with the 'Sephadex' G150 F2 fraction at a concentration of 100 µg in Freund's complete adjuvant $\times 2$; $\bigcirc$ — $\bigcirc$, F3 fraction at same concentration and schedule; Δ --- Δ , F1 fraction; Δ — Δ , Freund's adjuvant-inoculated controls. Challenge of 4.4×10^3 LSTRA cells was made 2 weeks after the last treatment. There were seven mice in each group, thus, maximum cumulative units of growth here is 21.

disseminates widely after intraperitoneal challenge with inoculations as low as 1×10^4 cells. CBF₁ mice pretreated with both the CM and F2 fraction preparations but not with F1 and F3 showed a striking resistance to FBL-3 challenge at cell concentrations of approximately the TD_{80-90} as reflected in frequency of leukaemic deaths. The difference of the mean frequencies of death in Fig. 4A was $70 \pm 23\%$ ($P < 0.002$) and in Fig. 4B, $62.5 \pm 16.2\%$ ($P < 0.001$) for the three groups combined, adjuvant control plus F1 and F3 fraction recipients versus the F2 fraction recipients. This activity of the F2 fractions observed in these *in vivo* studies correlated with the activity of F2 in inhibiting cytotoxicity of RBL-5 cells *in vitro*, as discussed below.

To test for the specificity of the immunogenic response to RBL-5 membrane antigen, C57Bl mice were pretreated with the CM preparation at the concentration and schedule in which resistance to leukaemic cell growth of LSTRA and FBL-3 was achieved. Challenge with syngeneic tumour No. 89 known to contain a polyoma virus-specific surface antigen¹⁶ did not reveal inhibition of the growth of this neoplasm (Table 1).

Antisera obtained from CBF₁ mice that received inoculations weekly for 3 weeks with 400 µg of the CM preparation were cytotoxic for ⁵¹Cr-labelled RBL-5 target cells in the presence of rabbit complement. This cytotoxicity titre increased from 1/32 to 1/128 (80% lysis) with continued immunizations over a further 3 weeks. These antisera did not neutralize RLV. Neither infectious RLV, as assayed in the indirect XC test¹⁷ nor type-C particles from electron microscopic study were observed in our CM preparation.

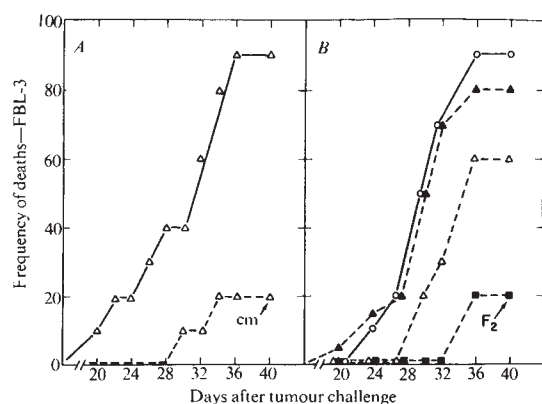


Fig. 4 Inhibition of growth of Friend virus-induced lymphocytic neoplasm, FBL-3, C57BL/1 male mice 12 weeks old. All challenge inoculations were made intraperitoneally from the ascites-maintained neoplasm 2 weeks after the last treatment subcutaneously with the RBL-5 antigen preparations. **A:** Δ — Δ , 400 μ g CM antigen in complete Freund's adjuvant inoculated subcutaneously $\times 2$; Δ — Δ , Freund's adjuvant-treated controls. Groups receiving 1×10^5 and 5×10^5 FBL-3 cells are combined. There were ten mice in each group. **B:** $\square$ — $\square$, mice treated with the F2 fraction, 100 μ g $\times 2$ at weekly interval; $\circ$ — $\circ$ and Δ — Δ , mice treated with the F3 and F1 fractions respectively, both at same concentration and schedule as F2; $\blacktriangle$ — $\blacktriangle$, Freund's adjuvant-treated controls. There were ten mice in each group and twenty in the control group. Results for groups receiving 5×10^5 and 1×10^6 FBL-3 cells are combined. The TD_{50} of FBL-3 was approximately 1×10^4 leukaemic cells. At 75 d after FBL-3 leukaemic cell challenge five out of ten (50%) of the F2 fraction-treated mice were alive and free of tumours, compared with only seven out of forty (17.5%) of the other combined groups.

In *in vitro* complement-dependent cytotoxic assays using RBL-5 target cells labelled with ^{51}Cr , inhibition of lysis was observed repeatedly using the CM preparation, the papain-solubilized CS and the pooled F2 fraction of antigen. A high-titred anti-MSV(MLV) antiserum and the antiserum produced in syngeneic mice after repeated CM immunizations, as discussed above, were used interchangeably. These antisera were not cytotoxic for normal lymph node cells of C57BL/6 origin nor for SV-AL/N fibroblasts known to contain SV40 VSTA¹⁸. The pooled F1 and the pooled F3 fractions at the same concentrations (4 to 10 mg protein ml^{-1}) and in identical conditions did not inhibit cytotoxicity.

We tried to calculate the overall yield of activity of this VSTA using inhibition of ^{51}Cr release (see ref. 18). If inhibition titres obtained with the CM antigen preparations are used (titres ranging from 1/64 to 1/128 at 50% inhibition of lysis), the F2 fraction contained 4 to 6% of the CM total activity. Activity was found, as assayed by inhibition of cytotoxicity, in the sediment and this is an indication that our papain diges-

tion has not destroyed the activity as measured *in vitro*; attempts are now underway to solubilize this remaining activity by methods other than papain digestion.

In the pooled F2 fraction as well as in the CM preparation, H-2 specificities 2 and 28 were also detected using monospecific alloantisera. The activity we observed in our cytotoxicity assays coincides with the pooled F2 fraction of H-2 specificities (see Fig. 5).

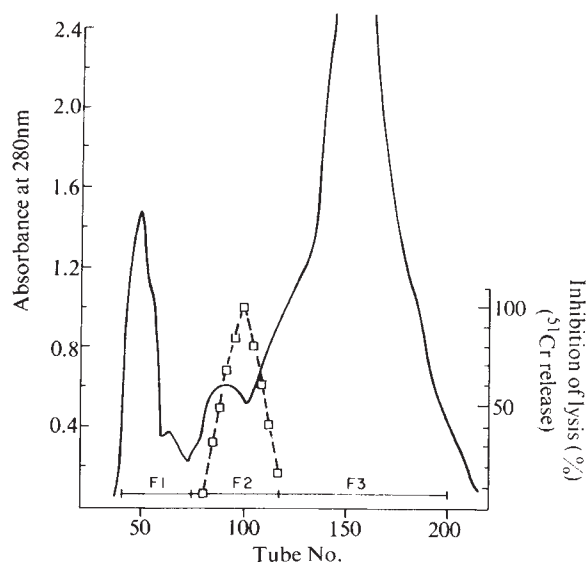


Fig. 5 Fractionation of papain-solubilized VSTA from 2×10^{10} RBL-5 ascitic cells on 'Sephadex' G150 column (2.3×150 cm) which was equilibrated with and eluted with 0.01 M Tris-HCl buffer, pH 8.2, and 0.15 M NaCl. —, Absorbance at 280 nm; $\square$ — $\square$, individual inhibitory activities of H-2.28 allo-antigenic specificity. The peak F2 when pooled at 4 mg ml^{-1} concentration was the only fraction containing inhibitory activity as assayed in the *in vitro* cytotoxicity test; peak F1 and F3 fractions were negative.

An obvious source of tumour antigens of the transplantation type in neoplasms induced by RNA viruses of the leukaemia-sarcoma complexes is infective virus or a structural part of the virus, since productive, non-lytic infection continues indefinitely in these induced neoplasms in contrast to neoplasms induced by DNA oncogenic viruses. Thus, expression of VSTA could be an expression of replicating, intact, infective virus incorporated in the cell membrane. Previous work from this laboratory¹⁹ and others^{20,21} using "non-releaser" neoplasms shows that VSTAs, at least of some RNA virus-induced tumours are of non-virion origin, but are nevertheless specified by the virus inducing the neoplasm.

Table 1 Test for the Specificity of RBL-5 Tumour Antigen C57BL Mice Immunized with RBL-5 Crude Membrane Antigen. Challenged with Polyoma-specific Neoplasm No. 89

Group	Treatment	No. polyoma tumour cells inoculated	No. with tumour No. in group	Mean tumour diameter (mm at days):			
				14	28	35	50
1	Crude membrane 400 μ g	5×10^4	5/5	7.2	8.3	12.3	16.2
		1×10^4	4/5	6.7	6.2	7.9	11.5
2	Freund's adj. only (Controls)	5×10^4	5/5	8.3	8.1	13.3	18.3
		1×10^4	4/5	7.4	6.6	6.8	11.4

Tumour No. 89 containing polyoma virus-specific surface antigen¹⁶ was inoculated subcutaneously into syngeneic C57BL/Ka female mice 14 d after last treatment with the crude membrane (CM) antigen preparation. The concentration of 1×10^4 tumour cells was determined in previous experiments to be approximately the TD_{50} . The mean tumour diameter is the arithmetic mean of length + width + depth. The time to death with neoplastic growths for the combined (5×10^4 and 1×10^4 groups) was 54.0 d for group 1 and 53.4 d for group 2. Tumour No. 89 failed to grow in 13 C57BL/Ka mice immunized with polyoma virus and inoculated with 2×10^4 dissociated No. 89 cells while nine out of fifteen (60%) untreated controls died of progressively growing tumours 56 d after grafting.

The tumour antigens isolated from RBL-5 discussed here, and responsible for the specific immunological reactions described seem also to be of nonviral origin. The CM material did not contain infective leukaemogenic virus as determined in the indirect XC assay, CM-immunized mice did not produce antiviral antibodies, as assayed in Rauscher-virus neutralization studies nor were any type-C particles observed in our CM preparations.

We conclude from these findings that a molecular entity with the characteristics of a tumour antigen of the trans-plantation type has been solubilized from a crude membrane preparation. Immunogenic properties of the VSTA have been preserved and this entity does not seem to be a virion antigen or a structural part of RLV. From the characteristic elution on 'Sephadex', this entity seems to be of relatively high molecular weight (about 50,000–60,000); it elutes in the same area in which both H-2 and non-H-2 activities have been shown to elute after solubilization with papain⁷.

As it is very unlikely that we are measuring an H-2 or non-H-2 activity (in our syngeneic system), the VSTA solubilized in these studies must represent a new surface component (antigenic determinant) that can be isolated and further analysed. This analysis we hope will shed some light on the mechanisms involved in the interaction between viral genes and cell regulating mechanisms.

We thank Donald Foor, Sam Morris and Thomas Fischetti for technical assistance. Dr Kenneth Chang performed the assays for infective RLV and for neutralizing antibodies to RLV; Dr Nelson Wivel performed the electron microscopic study for type-C particles. We thank Dr D. Lavrin for anti-MSV(MLV) antisera.

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Polyploidy and Flavonoid Synthesis in *Briza media* L.

POLYPLOIDY may increase the flexibility of a species and consequently its ecological range by extending physiological tolerance^{1,2}. In *Lycopersicon esculentum* polyploidy is reported to raise ascorbic acid levels in the fruit³ and in *Valeriana officinalis* the polyploids have been shown to be better adapted to certain soil types than the diploids⁴. Little work, however, has been carried out on chemical differences between chromosome races, so it was of some interest, when we found that diploid and tetraploid plants of *Briza media*, which have different geographical distributions also have distinct leaf flavonoid patterns. Diploid plants accumulate the C-glycosylflavones vitexin and isovitexin, tetraploid plants orientin and iso-orientin (Fig. 1).

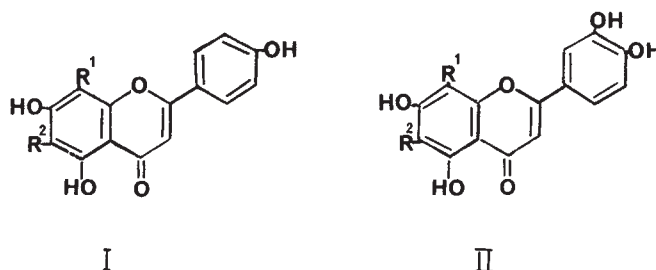


Fig. 1 Structures of flavonoids found in polyploids of *B. media*. I, vitexin, R¹=glucose, R²=H; isovitexin, R¹=H, R²=glucose. II, Orientin, R¹=glucose, R²=H; iso-orientin, R¹=H, R²=glucose.

Here we report on the leaf flavonoid patterns of artificially produced polyploids, differences in enzyme activity with ploidy level, the discovery of a new isomerase enzyme converting isovitexin to vitexin and possible effects of polyploidy on the natural distribution of *Briza media*.

The plants used were collected in the wild or grown from spontaneous seed. Artificial tetraploids were produced by colchicine treatment of seedlings and chromosome numbers obtained by root tip squashes⁵. Flavonoid identification has been described previously⁶. Hydroxylase activity was assayed by a modified version of the method of Vaughan and Butt⁷ using acetone powder enzyme extracts instead of a *brie* to remove interfering substrates present in the leaf. The powder was further purified by precipitation at 50% (fraction 1) and 100% (fraction 2) saturation with ammonium sulphate in 0.05 M citrate-phosphate buffer pH 5.4. Horizontal block electrophoresis of leaf and seed proteins was carried out on a 7.5% acrylamide gel, after the method of Lund⁸ as described by Crowden *et al.*⁹.

Leaf flavonoids have been examined in several artificially produced polyploids of *B. media*; triploids, formed by crossing naturally occurring tetraploids with diploids, artificial tetraploids and octoploids were all found to synthesize orientin isomers and indeed to have an identical chromatographic pattern with the naturally occurring tetraploid. Thus it seems that the addition of a single genome is sufficient to cause a change in flavonoid synthesis in *B. media* and the addition of more genomes has no further effect.

Vitexin and orientin are C-glycosylflavones differing in the number of hydroxyl groups on the B-ring (Fig. 1) but it is not known whether they are formed by consecutive steps in the same biosynthetic pathway¹⁰. There is evidence, however, that flavones (with no C-sugar) containing a 4'-mono-hydroxy B-ring can be oxidized to flavones with a 3',4'-dihydroxy B-ring in the families Lemnaceae¹¹ and Umbelliferae¹². C-Glycosylation appears to occur at a much earlier stage in biosynthesis than both O-glycosylation and O-methylation¹¹.

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Fig. 2 Distribution of diploid (●) and tetraploid (○) races of *Briza media* in Europe.

All attempts at isolating an enzyme from *B. media* for hydroxylating vitexin to orientin failed but during the course of these experiments a totally different enzyme activity was discovered. Thus isovitexin, when incubated at 37° C with the partially purified enzyme fractions at pH 5.4, was completely converted to vitexin in 1 h. The conversion was monitored between 0 and 6 h by paper chromatography using standard solvents¹⁰. Neither the reverse reaction nor the conversion of iso-orientin to orientin could be shown. Boiling for 20 min stopped the reaction completely in both diploid fractions and tetraploid fraction 2. Fraction 1 (tetraploid) remained active but was inactivated by 2 h boiling. Buffer controls (boiled and unboiled) showed no conversion of isovitexin to vitexin. The optimum pH for the reaction was around 6.8, at which conversion was almost instantaneous. At pH 2.6 the reaction was very slow and at pH's 4.0, 5.4 and 8.8 it was complete in 15 min (fraction 1) and 1 h (fraction 2), at pH 2.6–6.8 citrate-phosphate buffer and borate buffer pH 8.8. Neither vitexin nor the buffer controls showed conversion at any pH. It thus seems that the leaf of *B. media* contains an isomerase enzyme or enzymes converting isovitexin to vitexin and differing with ploidy level in heat stability. This is the first report of such an enzyme.

The function of the new enzyme in the plant is not yet clear, but it does not seem to have an important synthetic role, as the ratio of isovitexin to vitexin in leaf tissue is very much in favour of the 6-isomer, the ratio being approximately 4:1. Furthermore Wallace and Mabry¹³ have shown by tracer feeding experiments in *Lemna minor* that vitexin is converted to isovitexin *in vivo* but were unable to demonstrate the reverse reaction. The isomerase, therefore, may be more important in the metabolism and turnover of flavone C-glycosides and this is being further explored.

The proteins of diploid, triploid and tetraploid seeds were compared using horizontal block electrophoresis. Esterase and peroxidase activity was determined according to the method of Crowden *et al.*⁹. Esterase activity was found to be about four times greater in triploid and tetraploid leaves than diploid but peroxidase activity was equally strong in all three. Seed extracts were weak in esterase activity but the diploid gave two distinct bands, the tetraploid only one. The seed peroxidase reaction did not vary with ploidy but it was interesting that both cationic and anionic bands were observed.

It appears, therefore, that in *B. media* polyploidy affects some physiological processes such as flavonoid synthesis, esterase activity and the heat stability of the new C-glycosyl-flavone isomerase but does not affect peroxidase activity or the actual isomerase reaction. These physiological changes may be in some way enabling tetraploid races to extend the

range of the species by exploiting different climatic and edaphic zones. The geographic separations of the races (Fig. 2) would appear to support this idea. The tetraploids are most common in "central" Europe and have not been found in Britain or Scandinavia where all populations studied were diploid. The two races have only been found growing sympatrically in the Massif Central of France¹⁴—a botanically and geologically unusual area.

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Ethanol inhibits Intestinal Calcium Transport in Rats

ALTHOUGH it is known that intestinal calcium malabsorption may occur in chronic alcoholic patients indirectly as a result of steatorrhoea of multiple causes¹⁻⁷, producing inadequate absorption of fat-soluble vitamin D, little attention has been paid to possible direct effect on small intestinal transport. To test that possibility, I starved male albino rats 8 h before and 18 h after the acute administration of ethanol (7.5 g kg⁻¹ body weight) or isocaloric glucose (13.7 g kg⁻¹; they were, however, permitted free access to water. Chronic ethanol ingestion was achieved by giving 20% ethanol in water as a sole source of fluid and standard laboratory chow was fed *ad libitum* for 12 d. Comparisons were made with *ad libitum* and pair-fed controls. Calcium transport by duodenum and ileum was measured using an *in vitro* everted gut sac technique⁸.

The results of experiments measuring serosal/mucosal ⁴⁵Ca concentration ratios in animals given the single dose of ethanol (Fig. 1) indicate no significant difference in transport ratios ($P > 0.05$). In animals ingesting ethanol chronically there was no significant difference between the weights of pair-fed animals (124 ± 3 g) and ethanol-fed animals (124 ± 4 g) although both were statistically depressed compared with animals fed *ad libitum* (175 ± 5 g) ($P < 0.01$). Chronic administration of ethanol interfered with the ability of duodenum to transport calcium ($P < 0.01$) compared to *ad libitum* or pair-fed controls (Fig. 1). The differ-

ence between the two control groups is not statistically different. I found no differences in ileal concentration ratios ($P>0.05$). Examination of liver and intestines by light microscopy revealed normal histology in all groups and determinations of serum bilirubin, alkaline phosphatase and aspartate aminotransferase (GOT), which were performed to monitor hepatic function, were normal in the three groups.

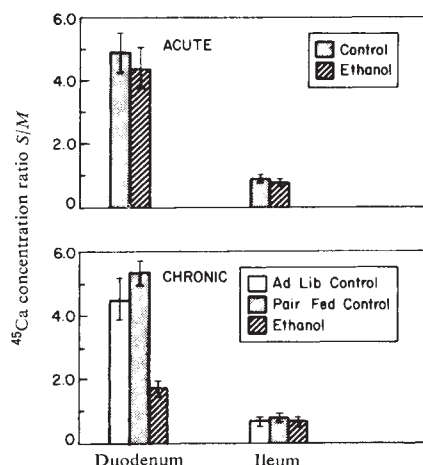


Fig. 1 Effect of ethanol *in vivo* on calcium transport by rat small intestine. Sacs 5 cm long, filled with 0.5 ml of a medium consisting of 0.125 M NaCl, 0.01 M fructose, 0.00025 M CaCl_2 , 0.030 M Tris-Cl, pH 7.4, and sufficient $^{45}\text{CaCl}_2$ to provide approximately 25,000 c.p.m. ml^{-1} , were incubated at 37°C, with continuous bubbling of 100% oxygen, for 90 min, in 25 ml Erlenmeyer flasks containing 10 ml of identical medium. Radioactivity was determined by liquid scintillation spectrometry. Data are expressed as a final concentration ratio of the tracer inside the sac (serosal medium) over that outside the sac (mucosal medium), S/M. Eight or nine animals were used in each group. Values represent mean standard error.

It seems that the chronic ingestion of ethanol directly inhibits active transport of calcium by the rat duodenum, that segment which has been shown to most avidly absorb calcium^{9,10}. Comparison to pair-fed malnourished controls indicates the effect is independent of partial starvation. No evidence of liver dysfunction or cholestasis was present as measured by histological appearance and liver function tests. Pancreatic dysfunction cannot be implicated in these studies because chronic pancreatitis appears only after the twentieth month of exposure¹¹. A direct toxic effect might be responsible for this direct interference, but the normal histology in these studies, as well as the normal appearance of duodenum and jejunum observed previously in humans¹²⁻¹⁵, indicate a lack of structural changes as evidenced by light microscopy. In rats ingesting ethanol for 9 to 12 months, however, ultrastructural changes in ileum have been recognized, in spite of normal light microscopic appearance¹⁶. The mechanisms underlying this inhibition of calcium transport and the clinical implications remain to be determined.

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The Regulation of Intestinal Calcium Transport by Vitamin D

THERE is much evidence that metabolites of vitamin D are fundamentally involved in calcium absorption by the intestine. Cholecalciferol (vitamin D₃) is hydroxylated at C₂₅ in the liver^{1,2} and then at C₁ in the kidney³⁻⁵. 1,25-dihydroxycholecalciferol is thought to be the active form, a hormone which promotes Ca transport by the intestine⁵⁻⁸. Ca translocation across the mucosal epithelial cell involves entry across the brush border and exit across the basal-lateral membrane. The site and manner of action of vitamin D metabolites has not been unequivocally established, but has been suggested to be at the basal-lateral membrane⁹, the brush border^{10,11}, or both^{12,13}. Kinetic studies in the normal rat led to the conclusion that entry across the brush border was rate-limiting for Ca translocation across the epithelium¹⁴. This would itself suggest that 1,25-dihydroxycholecalciferol regulated Ca absorption by acting at the brush border.

Male albino rats were fed from weaning on a vitamin D-deficient diet. This consisted of per kg, 528 g whole wheat flour, 192 g wheat germ, 48 g yeast extract, 48 g casein, 144 g sucrose, 18 g olive oil, 12 g CaCO_3 , 9.6 g NaCl, 10 mg ascorbic acid, 200 mg ferrous lactate, 200 mg MgSO_4 , 5 mg MnSO_4 , 5 mg CuSO_4 , 1 mg KI, 0.3 mg CoSO_4 and 3,000 I.U. vitamin A. The diet contained 0.52% Ca and 0.50% P.

Parsons has shown that everted duodenal sacs from rats fed this diet from weaning to six to eight weeks old showed no active transport of ^{45}Ca with mean serocal/mucosal concentration ratio = 1.2. Active transport occurred when the rats were dosed with 400 I.U. vitamin D: the mean ratio increased to 2.7 after 8 h and to 5.2 after 24 h (B. J. Parsons, personal communication).

In the first experiment six rats were fed the vitamin D-deficient diet for 3 weeks. Three of these were given 100 I.U. (2.5 μg , 6.5 nmol) cholecalciferol orally in olive oil; 48 h later 6 flasks of duodenal slices were prepared and incubated with 1 mM Ca (see legend to Fig. 1 for method). The Ca influx was $5.31 \pm 0.17 \mu\text{mol g}^{-1} \text{h}^{-1}$ (mean $\pm$ s.e. of mean). The mean influx for three rats given olive oil without cholecalciferol

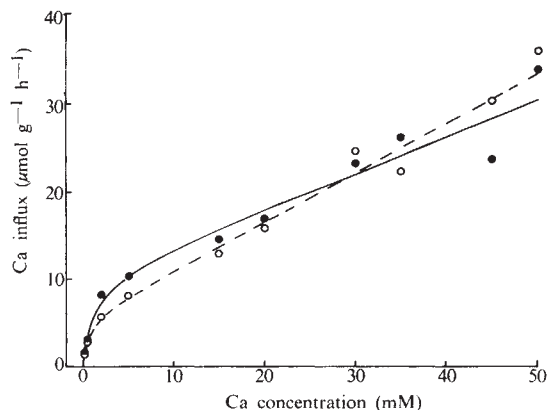


Fig. 1 Effect of cholecalciferol on Ca influx kinetics. Twelve rats were fed the vitamin D-deficient diet for 4 weeks. Six of these rats were given 100 I.U. cholecalciferol orally in olive oil, 6 given only olive oil. After 48 h duodenal slices were prepared and incubated as previously described¹⁴. Twelve duodenal slices were prepared from each rat, 1–5.5 cm from the pylorus. In each flask 6 slices, total fresh weight approx. 250 mg, were pre-incubated for 15 min in 8.1 ml. Ca-free medium containing 139.4 mM NaCl, 4.7 mM KCl, 4.0 mM Tris buffer at pH 7.4, and 27.8 mM glucose. Then 10.5 ml. medium was added with some Na replaced isomotically by Ca, labelled with ⁴⁵Ca. After 5 min further incubation the slices were washed twice with 154 mM NaCl, blotted and weighed. The uptake of ⁴⁵Ca was measured by liquid scintillation counting²³, to give an estimate of the unidirectional influx of Ca into the tissue¹⁴. Influx was across both mucosal and serosal surfaces. Each point gives influx for 1 flask of 6 slices. Points for 0.025 and 0.05 mM Ca are omitted. Curves were fitted for the expression given in the text. ●—●, Dosed with cholecalciferol; ○—○, vitamin D-deficient.

ciferol was $4.44 \pm 0.11 \mu\text{mol g}^{-1} \text{h}^{-1}$, which was significantly less by *t* test ($P < 0.01$).

Ca influx over a range of Ca concentrations has been analysed into saturable and linear components according to the expression¹⁴:

$$J = \frac{J_m[\text{Ca}]}{K_t + [\text{Ca}]} + P[\text{Ca}]$$

where J = Ca influx, $[\text{Ca}]$ = concentration, J_m = maximal value and K_t = half-saturation constant for the saturable (carrier) component, and P = constant for the linear (simple diffusion) component. This expression fitted the influx data well both in vitamin D deficiency and following 100 I.U. cholecalciferol in the second experiment (Fig. 1). There was a significant overall difference between the two conditions ($p = 0.016$): in particular cholecalciferol increased J_m from 5.51 ± 0.65 to $10.4 \pm 1.2 \mu\text{mol g}^{-1} \text{h}^{-1}$ ($p = 0.002$), increased K_t from 0.627 ± 0.096 to $1.22 \pm 0.18 \text{ mM}$ ($p = 0.008$), but decreased p from 0.556 ± 0.038 to $0.402 \pm 0.047 \text{ ml. g}^{-1} \text{h}^{-1}$ ($p = 0.020$). Except at high concentrations the overall effect was to increase Ca influx, and markedly so in the physiological range: the maximal increase was 38% at 3 mM Ca.

Thus the increase in Ca influx by cholecalciferol was the result of an increase specifically in the saturable component. This component is known to relate mainly to Ca entry across the mucosal brush border¹⁴, which was ascertained in normal, vitamin D-replete rats. The reduction in the saturable component induced by vitamin D deficiency must therefore indicate a reduction in that component of influx across the brush border. Martin and DeLuca¹¹ have also shown that cholecalciferol increased Ca influx across the brush border, but were unable to define Ca influx kinetics in vitamin D deficiency and did not identify a linear component. This analysis demonstrates that K_t was increased by cholecalciferol, from which it is unlikely that Ca influx was enhanced by the increased affinity of a carrier in the brush border. My results show for the first time that cholecalciferol facilitates Ca entry solely by increasing

J_m , which would indicate an increase in the number of entry sites in the brush border. The biochemical correlates of these could be (i) Ca-dependent ATPase¹⁵ (Ca entry is energy-dependent¹⁴), (ii) non-specific phosphatase^{15–17}, (iii) calcium binding protein¹³ or (iv) some other entity. As in the normal rat¹⁴, so with vitamin D action the kinetics of Ca influx correspond with the kinetics of Ca translocation across the entire epithelium: both *in vitro*¹⁸ and *in vivo*¹⁹ only the saturable component is increased by cholecalciferol.

In contrast, the linear component of Ca influx, P , was reduced significantly by cholecalciferol. This parameter is not specific for the mucosal surface¹⁴, but it is possible that physiological doses of the vitamin reduce Ca diffusion across the brush border.

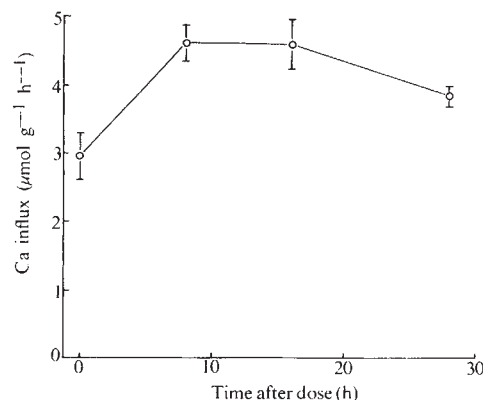


Fig. 2 Ca influx at intervals following administration of cholecalciferol. 16 rats were fed the vitamin D-deficient diet for 5 weeks. Groups of 4 rats were given 100 I.U. cholecalciferol orally in olive oil. After the times indicated 8 flasks of slices were prepared from each group and incubated with 1 mM Ca. Four control rats, shown at zero time, received no vitamin. Standard errors of the means are given.

The total Ca influx was determined at different times following the oral administration of 100 I.U. cholecalciferol (Fig. 2). The influx increased 56% by 8 h after the dose but was not further increased after 16 or 28 h. Both Ca absorption *in vivo*^{20,21} and transport across everted sacs (see above) had begun to increase by about 8 h after a similar oral dose. It would seem therefore that Ca influx across the brush border increases with vitamin D at least as early as these measures of Ca absorption. The time of increase in Ca influx also corresponds closely with the appearance of 1,25-dihydroxycholecalciferol in the mucosa following administration of cholecalciferol²².

My results support the conclusion that the primary functional response of the intestinal mucosa to 1,25-dihydroxycholecalciferol is to increase the number of sites of the carrier mechanism whereby Ca enters the cell across the brush border.

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Subcutaneous Growth of Human Tumours in Mice

CASTRO¹ reported the successful growth of human tumours in immune suppressed mice after the tumour tissue had been implanted beneath the left kidney capsule. In similar work we have used the subcutaneous route because we found this to be the most successful in our extensive investigations into tumour growth in ALS treated mice.

Female CBA mice were thymectomized at 4 weeks, whole body irradiated (900 R), 2 weeks later and reconstituted within 3–4 h of irradiation, using 1×10^7 syngeneic bone marrow cells, given i.v.

Table 1 Subcutaneous Tumour Growth

Tumour type	Percentage survival of tumours				
	1 wk	2 wk	3 wk	4 wk	5 wk
HeLa	100	100	100	53	21
Colon	100	100	100	80	
Stomach	100	100	50	38	38
Hep 2	75	75	50	50	12
Rectum	100	75	50	50	
Caecum	100	100	50	50	25
Breast adenocarcinoma	0	0	0	0	0
Secondary tumour, the primary being in the breast	70	70	70	70	
Lymph node from melanoma	100	100			

The tumours were obtained at operation and transported to the laboratory in Eagles minimal essential medium, containing 2% calf serum, 200 units of penicillin, and 100 mg streptomycin. Tumour pieces of about 2 mm were implanted subcutaneously in a ventro/lateral site over the right hip joint. In our ALS experiments we found by experience that this was the most suitable site for implantation, in view of the large tumour growths obtained, often in excess of 2–3 cm. In our present investigations we are able to report colonic tumour growths of the order of 12 mm within eleven days of implantation. This was assessed by direct palpation. Tumour types investigated are given in Table 1.

Our results clearly show that the subcutaneous route pro-

duces good tumour growth which is easy to assess by direct palpation and therefore affords a means of judging the effectiveness of treatment without having to kill the mice. We feel this method will be useful for further *in vivo* investigations of human tumours.

We thank New End Hospital and Hampstead General Hospital for the supply of some of the tumours.

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Inhibition of Pupariation in *Sarcophaga bullata*

THE classical test used as the bioassay for the insect moulting hormone, ecdysone, is based on the failure of the posterior part of ligatured larvae of *Calliphora erythrocephala* (or other fly species) to tan unless an adequate dose of ecdysone is administered. It is crucial for the success of the test to select larvae at the right stage of development, and as closely as possible to the critical time for pupariation¹. Larvae ligatured before the critical stage of natural ecdysone release may eventually pupariate in the front part where the hormone originates, but will never do so in the hind part. Post-critically ligatured larvae have had sufficient ecdysone released into the haemolymph previous to ligature to induce tanning in the hind parts. It is evident that at this time the anterior parts must also be post-critical. In larvae ligatured at this stage the anterior parts usually contract to the puparium and tan whereas the posterior parts merely tan without change in shape. The occurrence of tanning in the posterior part alone is therefore a paradox. Fraenkel² along with the description of the "*Calliphora* assay" also reported that ligatured larvae occasionally tanned only in the posterior region. Several investigators have been subsequently surprised by this phenomenon of "anterior inhibition", but there has been no agreement as to its cause.

Kaplanis *et al.*³ observed anterior inhibition in the larvae of *Musca domestica*. Langley⁴ attributed anterior inhibition in the larvae of *Glossina morsitans* to an inhibitory influence of the central nervous system which in a ligatured larva is confined to the anterior part. In contrast, Price⁵ hypothesized the presence of an inhibitory factor of the phenol oxidase inhibitor type, to account for a temporary inhibition of pupariation in the anterior parts of *C. erythrocephala* larvae ligatured between segments 8 and 9. Fraenkel and Zdarek¹ reported that in different species of flies the percentage of anterior inhibition is different. In the study reported here, with *Sarcophaga bullata*, it varied, according to the condition of the experiment, between 0 and 80%. Chang⁶ observed a 96.9% anterior inhibition in *Lespesia archippivora* that had been in his view ligatured "pre-critically", and suggested that, contrary to common notions, ecdysone was produced in the posterior parts only. The injection of ecdysone into such anterior parts did not, however, induce tanning. In the absence of evidence about the pre-critical nature of his larvae we suggest that they were indeed ligatured post-critically and showed the same effect of anterior inhibition observed in the other species.

Our studies now confirm Fraenkel's original view² that the inhibition of pupariation in post-critically ligatured larvae was

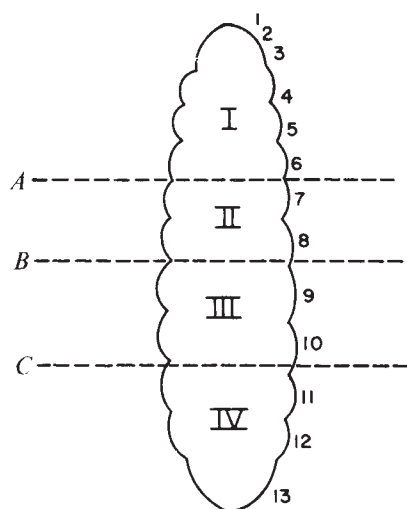


Fig. 1 Diagrammatic figure of larva of *S. bullata* showing positions of ligature.

caused by lack of oxygen. The experiments described here were carried out with *S. bullata* larvae, in the post-critical state, about 2–3 h before pupariation, when the region around the spiracles has begun to tan.

The anterior inhibition of post-critically ligatured larvae could conceivably be due to the inactivation of ecdysone⁷. Karlson and Bode⁸ reported the presence of such an inactivating enzyme in *C. erythrocephala* larvae. The rate of inactivation in whole larvae was greater than that in posterior parts of pre-critically ligatured larvae. Does the anterior inhibition result from the more rapid inactivation of ecdysone in the anterior parts? Massive doses of β -ecdysone (1 and 2 μ g, that is, 30 to 60 *Sarcophaga* units⁹) injected into each of fifteen inhibited anterior ends in each concentration within 1 h of tanning of the posterior ends, did not result in the subsequent tanning in any of them, ruling out the inactivation of ecdysone as the probable cause.

Langley's hypothesis⁴ of the central nervous system inhibiting the action of ecdysone in response to stimulation of surface sense organs was not supported by any evidence. Tetrodo-

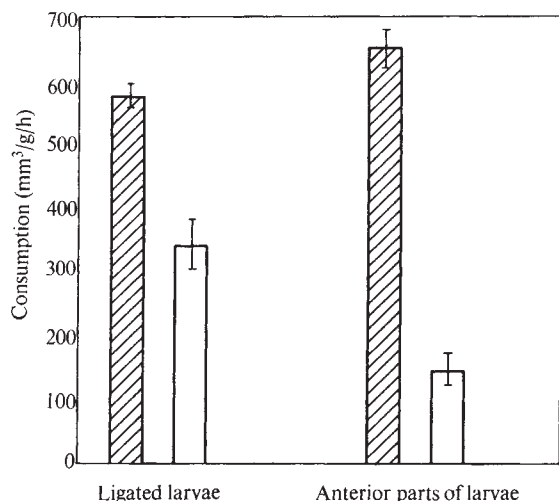


Fig. 2 Comparison of oxygen consumption in anteriorly inhibited and uninhibited whole ligatured larvae and isolated anterior parts. Bars indicate one standard error on either side of the mean value. ■, Uninhibited (tanned); □, anteriorly inhibited (untanned).

toxin had been shown by Zdarek and Fraenkel^{10,11} to cause complete paralysis and a complete or substantial block of nerve conduction without blocking pupariation in *S. bullata* larvae (0.1 μ g per larva or considerably less). The injection of 0.05 μ g of tetrodotoxin into anteriorly inhibited ends did not release the inhibition of pupariation, suggesting the absence of a significant inhibitory stimulus from the sense organs to the central nervous system.

Price⁵ observed that in *C. erythrocephala* larvae ligatured between segments 6 and 7, 8 and 9, and 11 and 12, segments 7 and 8 alone remained untanned. This he attributed to the presence of an inhibitory factor, of a phenol oxidase inhibitor type, in this region. In repeating Price's experiments, with *S. bullata*, the ligatures were made so as to minimize injury to the larvae, that is, not too tightly. In the first experiment the larvae

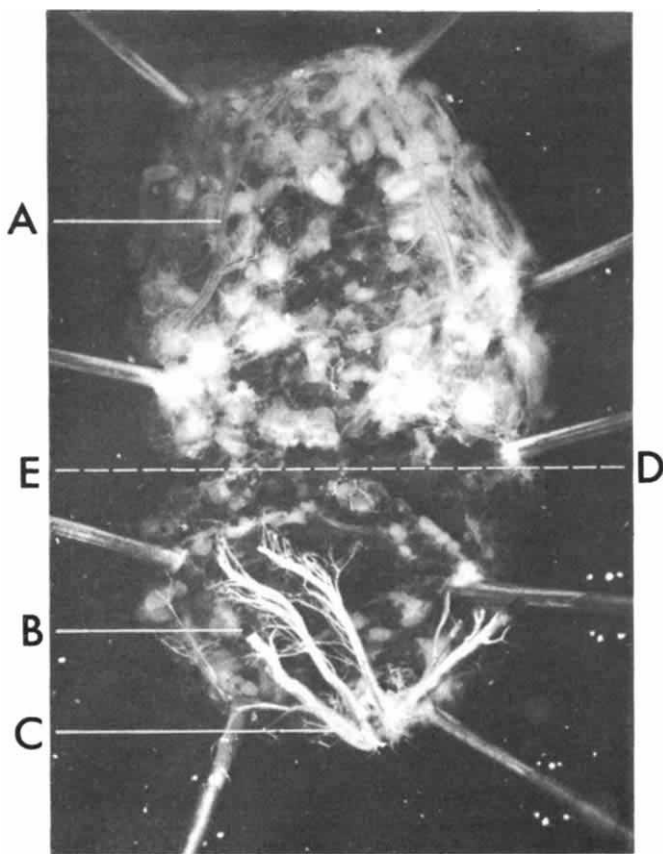


Fig. 3 Dissection of a ligated larva, exhibiting anterior inhibition of pupariation. A, Tracheae filled with haemolymph; B, plug of coagulated haemolymph; C, air-filled tracheae; D—E, position of ligature.

were ligatured at positions A, B and C (Fig. 1) in that order. The results were identical with those of Price; only region II remained untanned when observed 6 h and 25 h later. However, when the ligatures were applied in the reverse order of C, B, A, only region III remained untanned. When ligatured in the order A, C, B, or C, A, B, both regions II and III remained untanned. Each of these sets of experiments was carried out with twenty-five larvae, with invariably the same result. Thus the inhibition always occurred between the first two ligatures applied irrespective of their positions. In no case did regions I and IV, which bear the spiracles, show inhibition. (The expression of anterior inhibition is variable.) We found that the first two ligatures can be tightened very firmly without rupturing the body wall, whereas if the third ligature was tightened to the same degree it would cause a rupture with haemolymph flowing out. Thus the third ligature was never applied as tightly as the first two, and

did not cause a disconnection of the tracheal trunks. The invariable tanning of the two regions bearing the spiracles and the inhibition of the spiracle-less regions of the larvae between the first two ligatures to be applied indicates that inhibition is caused by the cutting off of the tracheal supply of the air through the spiracles. These experiments were repeated with *C. erythrocephala*, with the same results. Therefore the need to postulate an inhibitory factor, localized in a particular region, does not arise.

If the inhibition is caused by the cutting off of the tracheal supply of air, it would follow that the larvae showing inhibition would consume less oxygen than those that do not show it. Oxygen consumption was measured with a Scholander respirometer (Mark Co.), at 26° C. Measurements were made at hourly intervals for 4 h from the time of ligation, and the volume of oxygen consumed per hour per gram wet weight was computed. Where the oxygen consumed by the anterior parts only was measured, the posterior parts were cut off after ligation between segments 8 and 9. The results (Fig. 2) are mean values for at least ten specimens tested separately. Inhibition in a particular larva cannot be predicted at the time of ligation and can be observed only 2–3 h later.

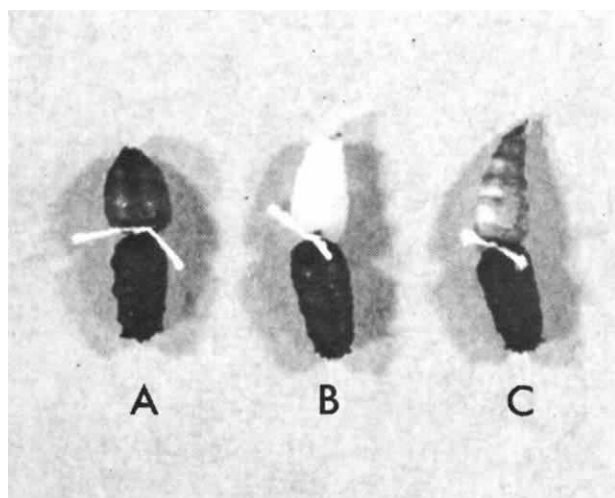


Fig. 4 Effect of oxygen on anterior inhibition of tanning. A, Ligatured larva with no anterior inhibition; B, ligatured larva with anterior inhibition; C, ligatured larva with anterior inhibition, subsequently subjected to a stream of oxygen.

The volume of oxygen consumed by the ligatured larvae which subsequently showed anterior inhibition was approximately 70% of that of larvae that did not show it. This is because of the decreased uptake of oxygen by the inhibited anterior ends. Inhibited isolated anterior ends consume only about 25% of the oxygen consumed by the uninhibited anterior ends (Fig. 2). This reduction in oxygen uptake closely resembles that in sections of *C. erythrocephala* without spiracles¹².

Our experiments have shown that spiracle-less regions firmly isolated by ligatures do not tan. The measures of oxygen consumption showed a startling decrease in anterior ends destined to be inhibited, but these anterior parts bear the prothoracic spiracles. In *S. bullata* larvae, like those of many fly species, only the prothoracic and the last abdominal spiracles are functional. The anterior spiracles are, however, far smaller. Is the decreased oxygen consumption a direct result of the cutting off of the tracheal air supply in these cases too, and if so, how is it caused?

Dissection of anterior parts showed that internal wounding caused the inhibition. The tracheae in the inhibited specimens were completely severed at the point of ligation and did not appear silvery, but instead were filled with a liquid (Fig. 3). They appeared colourless at the time of dissection, and gradually acquired a black colour reminiscent of the darkening of haemolymph when exposed to air. The tracheae of the uninhibited anterior ends remained silvery due to the air within the lumen. Here too the tracheae were severed in some, however the severed ends were covered with a black plug of tissue (as seen in the posterior part of Fig. 3). Thus it seems that the decreased oxygen consumption in the inhibited anterior ends comes about because the tracheal lumina is flooded with haemolymph, thus preventing air from reaching the tissues.

To confirm this view, anteriorly inhibited larvae in which the tracheae are blocked were subjected to a steady stream of oxygen for 6 h. These anterior ends which normally never tan showed tanning to a certain degree after this treatment (Fig. 4). This shows that the higher oxygen uptake allowed some tanning to take place.

Thus, inhibition of tanning in post-critically ligatured fly larvae can be produced either by eliminating the spiracles, or by abolishing tracheal function as a result of the spiracles being filled with liquid. It is interesting that in tsetse flies the anterior spiracles are not open to air which readily explains the 100% inhibition⁴ of tanning in the anterior parts. It is known that tanning of the puparium depends on oxygen reaching it from inside through the tracheae, since too little penetrates through the cuticle from outside^{2,12}. The situation is changed radically in pure oxygen when oxygen consumption in the absence of spiracles is at least three times that in air¹² and sufficient oxygen can penetrate to the sites of action in the anteriorly inhibited specimens to cause considerable tanning. Anterior inhibition therefore results from a lack of oxygen caused by the injury to the tracheal system during ligation. The extent of this injury depends on the tightness of the ligation, and is different in different species.

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Tension Receptor Reflexes in the Walking Legs of the Crab *Cancer pagurus*

THE decapod crustaceans have many advantages for the study of the neuronal mechanisms controlling simple behaviour¹⁻³. Numerous authors have investigated the functions of the walking legs, but models of the control mechanisms have been presented in terms of central programmes and reflexes related simply to the positions and movements of the joints^{4,5}; little attention has been paid to the control of tension in the chief working muscles. It has been shown, however, that the tension of some of these muscles is monitored by sense organs⁶. We present here some initial findings on the reflexes produced by input from these tension receptors and the integration of such reflexes in the intact animal.

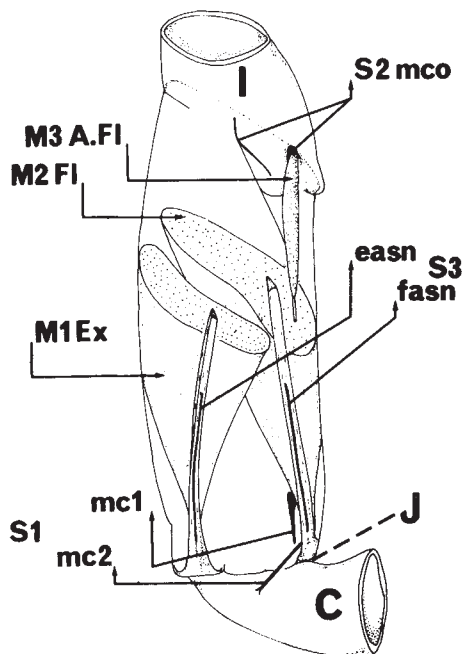


Fig. 1 The organization of the muscle (M1-3) and sensory (S1-3) systems controlling the M-C joint (J) of the first right walking leg. All the structures lie in the merus which is seen from the anterior surface lying between the ischium (I) proximally and the carpus (C) distally. The extensor (M1 Ex) lies on the anterior side of the segment and attaches to the carpus dorsally. It is innervated by two excitatory neurones and one inhibitory motor neurone. The flexor (M2 FI) lies on the posterior side of the segment and attaches ventrally to the carpus. It is innervated by four excitatory neurones and one inhibitory motor neurone. Further, a small accessory flexor muscle (M3 A.FI) attaches by way of a tendon to the distal end of the flexor apodeme. The simple chordotonal organs (S1 mc1, mc2) lie ventrally near the joint and are attached to the flexor muscles. For clarity the distal part of the accessory flexor muscle and the sub-groups of the myochordotonal organ (S2 mco) are not shown.

The investigation was carried out on the merus carpus (M-C) joint of *Cancer pagurus*. The organization of this joint (Fig. 1) is very similar to that of *Cancer magister*⁷. It allows almost 180° of movement in the dorso-ventral plane and is controlled by two major muscles which lie in the merus. Three types of sense organ are known at M-C. The simple chordotonal (S1) organs monitor joint position and movement, and generate resistance reflexes which oppose imposed (passive) joint movements⁸. It is curious therefore, that these reflexes should operate during walking to oppose ongoing (active) movements. In the propus-dactylus joint resistance reflexes do not occur

during undisturbed walking on even surfaces (that is, where the possibility of imposed load variations from cycle to cycle has been eliminated), but the mechanism governing this inhibition is unknown⁴. At M-C it is thought that in such circumstances the resistance reflexes operate only at extreme joint positions⁵. The complex myochordotonal organ (S2) monitors joint movements but it can also be set, without joint movement, by output to the accessory flexor muscle from the CNS. During movement the receptor generates subsidiary resistance reflexes. Its principal role, however, is thought to be in a complex set of reflexes which stabilize joint resting position⁵.

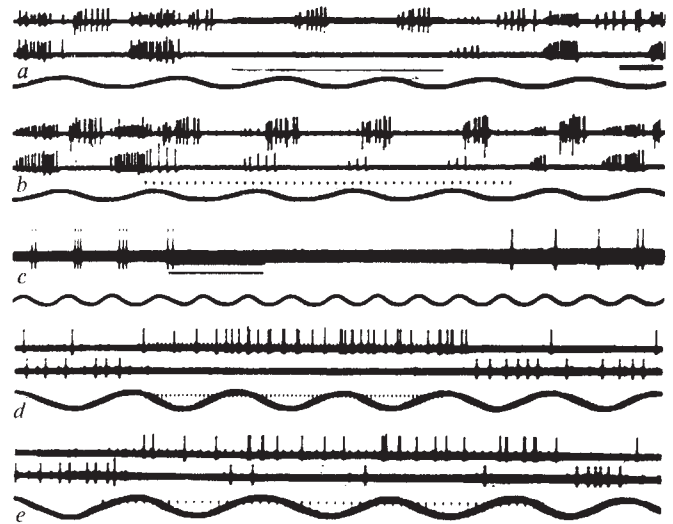


Fig. 2 Effects of stimulations of apodeme sensory nerves on resistance reflex motor activity. All stimulations were series of pulses of length 0.5 ms at low stimulus strengths. Each set of traces is described from top to bottom. *a*, Flexor plus extensor myogram, extensor myogram, stimulus of 50 pulses s⁻¹ (line), movement of merus-carpus joint with flexion, an upward deflexion and extension downwards; *b*, as in *a*, but stimulus 5 pulses s⁻¹; *c*, extensor motor nerve, stimulation at 20 pulses s⁻¹, joint movement with flexion downwards; *d*, extensor myogram, flexor myogram, joint movement with extension upwards, stimulation at 20 pulses s⁻¹; *e*, as in *d*, but 10 pulses s⁻¹. Time mark is 1 s in *a* to *c* and 0.5 s in *d* to *e*.

The tension receptors (S3) are associated with the extensor and main flexor apodemes. The sensory fibres run with the respective motor nerves proximally but separate distally to form distinct apodeme sensory nerves. Each apodeme sensory nerve carries numerous axons from bipolar cells whose dendrites run on the surface of the apodemes. Little activity was recorded in either sensory nerve when the joint was moved passively with low tension in the relevant major muscles. When the joint was fixed and the motor supply to a muscle stimulated, sensory responses were recorded from its apodeme sensory nerve at tensions well within the normal range observed in intact animals. All the units sampled responded with an increase in activity to increasing tension but other responses could be present in small neurones⁶.

In our experiments crabs were placed in a clamp with a platform on which the merus of the first right walking leg was fixed with the anterior surface exposed for dissection (the levator muscle apodeme of the leg was cut to prevent autotomy). Myograms were recorded from both major muscles with implanted insulated copper wire electrodes, and after a simple dissection, platinum wire electrodes were used to stimulate the extensor apodeme sensory nerve and to record from branches of the extensor motor nerve. Alternatively, by dissecting further into the meropodite the platinum wire electrodes could be used to stimulate the flexor apodeme sensory nerve and to record from branches of both flexor and extensor motor nerves.

Movements of the carpus were made with a potentiometer-monitored micromanipulator. Conventional methods of stimulation, amplification, display and photography were used.

Stimulation of the extensor apodeme sensory nerve at very low voltage (that is, activation of only large receptor neurones) effectively inhibited any spontaneous extensor motor activity. Extensor motor activity produced by evoking resistance reflexes (initial part, Fig. 2a) was also inhibited by stimulation of the extensor sensory nerve. Total inhibition was always obtained at 50 pulses s^{-1} (Fig. 2a), but the inhibition could be reduced by lowering the stimulus frequency (Fig. 2b). After a stimulus, inhibition could persist for many seconds (Fig. 2c). The effects on the antagonistic flexor motor neurones were more varied. In some examples there was little change in flexor activity during stimulation of the extensor sensory nerve (Fig. 2a, b) but more often an excitation was observed. We suspect that other factors (for example, the flexor apodeme sensory input) are involved in the antagonistic motor response. Stimulation of the flexor apodeme sensory nerve during passive movements of the carpus produced effects symmetrical to those observed for the extensor sensory nerve. At low stimulus strengths inhibition of the flexor motor activity was obtained (Fig. 2d) and this effect could be modulated by changes in stimulus frequency (Fig. 2e). Again, more variable results were obtained for the antagonistic (extensor) motor neurones but most often an excitation was observed (Fig. 2d, e). No significant changes in any of the responses to low stimulus strength activation of the apodeme sensory nerves were observed after destroying the myochordotonal organ receptor system by removal of the accessory flexor muscle.

Stimulation of either apodeme sensory nerve at much higher stimulus strengths produced, after a short latency, a strong discharge in both extensor and flexor motor neurones. In all experiments the identities of the stimulated and recorded nerves were subsequently verified by staining with methylene blue. Additionally, a series of control experiments confirmed that the results were obtained from apodeme sensory input to the CNS.

The crab M-C joint has now been shown to have three different types of sensory receptor, each of which can be divided into at least two distinct sense organs. In view of this complexity findings of no malfunction after ablation of single sense organs in such situations do not necessarily mean that the ablated receptor has no function in the intact animal^{9,10}. The widespread occurrence of tension receptors in the decapods⁶, and the possibility of hysteresis in the responses of crustacean muscles to motor output¹¹, also mean that for this group the regulation of muscle tension can no longer be ignored in models of neuronal control mechanisms governing simple behaviour.

The fact that the apodeme sensory input, which is dependent on muscle tension, can profoundly modify the resistance reflexes due to the chordotonal organs, may explain the inhibition of resistance reflexes during undisturbed walking on even surfaces, although an entirely central inhibition would seem to be a more economical mechanism. We suggest, however, that during normal walking on uneven surfaces the crab is as likely to encounter resistance to active movements as imposed (passive) joint movements. Thus reflexes generated by tension receptors (when an active movement is prevented) could be just as important as resistance reflexes in the intact animal. Tension receptor reflexes are clearly of some importance in cockroach walking¹².

The demonstration of a negative feedback loop from sense organs in series with the meropodite working muscles suggests that the apodeme receptors are analogous to vertebrate Golgi tendon organs. We would be cautious of a direct analogy because the effects of higher stimulus strength activation of the sensory nerves suggest that the apodeme sense organs also contain elements which evoke strong excitatory reflex output to the innervated muscle. More sophisticated experiments will be required to determine the nature of any such positive feedback

elements. The same holds for the more complex effects that we have observed for the antagonistic muscles in these experiments. Our findings nevertheless show that the sensory array at the crab M-C joint is more comparable to the vertebrate system than was previously thought, and thus probably more useful for the study of general principles.

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Trend towards Earlier Menarche in London, Oslo, Copenhagen, the Netherlands and Hungary

Brundtland and Walløe¹ have reported that the trend toward earlier maturation in children, as evidenced by age at menarche, has stopped, at least for the present. Using probit estimates in a *status quo* inquiry they gave the mean ages as 13.27 yr in 1952 and 13.24 in 1970. Both results were based on large samples of Oslo schoolgirls.

Here I report a similar finding in London schoolgirls. In a large-scale survey² made in 1959 in randomly selected London schools the mean, estimated by probits, was 13.05 ± 0.02 yr. A survey made between November 1966 and February 1967 (unpublished data), using a similar sample, gives a mean of 13.02 ± 0.03 yr. This figure relates only to those girls (approximately 7,000) between the ages of 9 and 16 whom the health visitor questioners had noted as "European" as opposed to "non-European". Because there were fewer non-Europeans in 1958 than in 1966 this may make the two samples more comparable. Inclusion of non-European girls, however, alters the value only trivially.

The previously well marked trend, therefore, seems to have stopped in London and Oslo. This is not (? yet) the case in all parts of Europe. Two other studies, comparable in both numbers and method with those cited, show that in Holland³ between 1955 and 1965 the mean age decreased from 13.6 yr to 13.4 yr, and in West Hungary⁴ between 1960 and 1965 it

decreased from 13.27 ± 0.07 to 13.13 ± 0.01 yr. A slightly less reliable sample of Copenhagen girls showed a decrease from 13.8 ± 0.03 (ref. 5) in 1950 to 13.2 ± 0.2 in 1964 (ref. 6). Some of the best series are illustrated in Fig. 1).

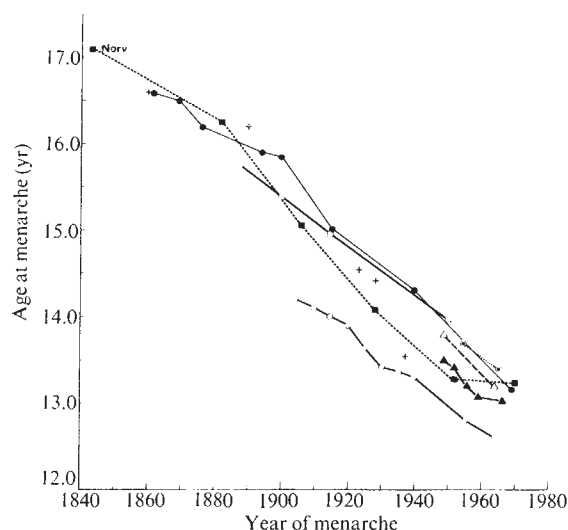


Fig. 1 Trend in menarcheal age in some European countries and the United States 1840–1970. Redrawn, with additions, from Tanner⁷. Δ , Denmark; $\bullet$, Finland; +, Germany; $\circ$, Holland; $\blacksquare$, Norway; $\square$, Sweden; $\blacktriangle$, United Kingdom; $\circ$, United States.

I suggested earlier that the trend would continue until North European means reached somewhere near the value for Americans⁸ (about 12.7 yr), Italians⁹ (about 12.6 yr), or even perhaps Chinese^{10,11} growing up in economically similar circumstances (about 12.4 yr). It looks as if this expectation will prove to be wrong; in the next decade Dutch, Danes and Hungarians may join the standstill of the Atlantic fringe.

The data on London girls will be published in detail elsewhere¹². I acknowledge the initiative and cooperation of the Inner London Education Authority and the support of the Nuffield Foundation.

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Outbreeding and Inbreeding in a Zinc-Lead Mine Population of *Armeria maritima*

ANTONOVICS¹ and Lefèbvre² have shown that self-fertility occurs in zinc-lead mine populations of normally incompatible species (*Agrostis tenuis*, *Anthoxanthum odoratum*, *Armeria maritima*). In *Agrostis* and *Anthoxanthum*¹ this change to inbreeding is suggested to be an adaptation to reduce gene flow from plants growing adjacent to the mine that do not tolerate metals. In *Armeria*² there is no surrounding non-tolerant population and self-fertility has been assumed to be a mechanism usually found in colonizing species which enables a reliable production of seeds in colonizing situations where individuals are both few and sparse^{3,4}.

Table 1 Number of A-types and B-types in Ten Samples Collected from the Population of Plombières

Samples	A-type	B-type	$\chi^2_{50:50}$	$\chi^2_{48:52}$
1	461	383	5.54	12.49
2	127	123	0.06	0.79
3	138	122	0.98	2.60
4	272	267	0.02	1.25
5	194	201	0.16	0.16
6	147	181	3.64	1.22
7	47	53	0.36	0.04
8	46	54	0.64	0.16
9	54	46	0.64	1.44
10	51	49	0.04	0.36
Total	1,527	1,479	12.08	20.51

In these experiments inbreeding was artificially induced by enclosing inflorescences in paper bags. This method gives data on potential self-fertility but does not give any idea about the efficacy of self-fertility in the mine population. Selfing obtained by artificial methods can disappear in natural conditions⁵. I report here results on outbreeding estimates established for a population of *Armeria* collected from the mine site of Plombières (Belgium). Mating-type genes were used. Reproduction, in European populations of *Armeria*, is controlled by a dimorphic self-incompatibility system which breaks down to monomorphic self-compatibility in Arctic and New World races⁶. In European populations, half the individuals are heterozygous with reticulate pollen and smooth stigmas (A-type AC/ac) and the other half are homozygous with punctate pollen and papillous stigmas (B-type, ac/ac). Pollen type and stigma type are determined by the supergene AC (ref. 6). Fertile combinations are only possible between A-type and B-type so that the 50:50 ratio is maintained through generations.

Table 2 Statistical Analysis of Data in Table 1

Source	d.f.	$\chi^2_{50:50}$	$\chi^2_{48:52}$
Total	10	12.08	20.51*
Pooled	1	0.69	9.65†
Interaction	9	11.39	10.86

* $P < 0.05$. † $P < 0.01$.

If inbreeding occurs in a 50A/50B population there will be a predominance of B-type, as found by Baker is a population collected in the Shetland Islands, in which some individuals were self-fertile.

Table 3 Results of Progeny Tests in Three Samples of the Population of Plombières

Samples	Plombières 1		Plombières 2		Plombières 3		Total	
Parents	B-type	A-type	B-type	A-type	B-type	A-type	B-type	A-type
Number of parents tested	18	8	14	19	19	15	51	42
Offspring B	383	123	122	267	201	181	706	571
Offspring A	451	127	138	272	194	147	783	546
Frequency of B	0.460	0.492	0.470	0.495	0.509	0.551	0.473	0.511
Frequency of A	0.540	0.508	0.530	0.505	0.491	0.449	0.527	0.489
Outbreeding coefficient (λ)	1.024		1.020		1.093		1.049	

If all the individuals in an initial 50A/50B population become completely self-fertile a new equilibrium will be established after a few generations at 25A/75B. As the mean self-fertility in the Plombières population is approximately 7%, a 48A/52B ratio must theoretically be reached.

To test this disequilibrium, population samples were scored for A and B types (Tables 1 and 2). Statistical analysis shows clearly that there is no significant deviation from the 50A/50B ratio and that the results are statistically incompatible with a 48A/52B ratio. This suggests that inbreeding is not occurring at the present time in the population.

In addition, progeny tests were carried out on three samples of the population (Table 3) to estimate the outbreeding coefficient according to the method of Vasek⁷. Two equations are solved:

$$\lambda p = DR \quad (1)$$

frequency of dominants in the progeny of recessive homozygous.

$$\frac{1}{4}(1 + \lambda q - \lambda p) = RH \quad (2)$$

frequency of recessives in the progeny of heterozygous where λ =frequency of outcrossing, p =frequency of dominant gene, q =frequency of recessive gene.

Because of the breeding system in *Armeria*, where $DR=0.50$ and $p=0.25$, λ has a theoretical value of 2; this value must be divided by two as only half of the possible crosses exist. Table 3 shows that in the three plots λ has a value of approximately 1.

On the other hand, if inbreeding occurs in the population, some A plants should be homozygous dominant (AA) and produce only A-type progeny in the progeny test. If total inbreeding occurs, the frequency of AA in the A-type progeny will be 0.50. The potential amount of inbreeding in the population (7%) leads to the estimate that approximately 4% of AA plants must arise in the population. In our experiment all the A-plants tested give A and B-types, without exception, in their progeny. This is additional evidence against self-fertility in this population.

It is thus obvious that outbreeding is complete at the present time in the mine population of Plombières although some potential self-fertility has been shown artificially.

The adaptive significance of self-fertility in mine populations must therefore be reviewed. In Plombières nearly all plants grow at a high density so that there is no spatial barrier to outbreeding. It is also probable that pollen competition favours foreign pollen. In this situation self-fertility has no adaptive value. But if new areas on the mine are available for colonization, self-fertility could be important. To test this assumption progeny tests need to be carried out on plants isolated during a colonization.

In fact, self-fertility seems to be the relic of an adaptive character which has no effect now in the dense mine population studied here. In *Agrostis* and *Anthoxanthum* more information is needed on the efficacy of self-fertility in the mine population compared with gene flow from the sur-

rounding population because self-fertility could be just an adaptation to initial colonizing stage as in the case of *Armeria*.

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New Endocranial Values for the East African Early Hominids

THIS is a preliminary report on research carried out during 1971 and 1972 on the endocranial values of some East African hominids from Olduvai Gorge, Tanzania, and East Lake Rudolf, Kenya. The cranial remains suitable for the reconstruction of volumetrically accurate endocranial casts are as follows: from Olduvai Gorge, hominids OH 7, 9, 12, 13 and 24; from East Lake Rudolf, KNM-ER 732 and 406. Each set of hominid remains required its own reconstructive procedures; a full description of each is unnecessary here but will be published elsewhere. In this communication I report only the results and make some preliminary observations as to their significance. Table 1 shows the cranial capacities found for each hominid. Not all of these results should be equally regarded; some results are only approximations but others are stated with more confidence.

The cranial capacities for OH 12, 13 and 24, are based on partial plasticine reconstructions added to accurate plaster casts of the interior of the cranial fragments. OH 12 and 13 required considerable addition of plasticine, particularly in the frontal region.

OH 24 required correction of the distortion of the existing endocranial cast as produced by Mr Ron Clarke. Two coronal sections were made in a plaster cast of the original reconstruction as most of the distortion seemed to be in the vertical plane. The sections roughly divided the endocranial into thirds which were then rotated so that the dorsal segments left a small gap on either side of the central portion. Some small degree of reduction of the biparietal width was necessary for the distortion has clearly enlarged this dimension. Finally, the endocranial of the brain stem was carved out with a diamond rotary saw, and was correctly oriented in a more posterior position.

All of the reconstructions except KNM-ER 406 were coated with preservative and immersed in water to give the final displacement volume¹. OH 7, based on the juvenile parietals, and estimated at 687 cm³ by Tobias^{2,3} was not reconstructed, but "fitted" over existing full endocasts. The endocast of OH 16 was not reconstructed as the skull reconstruction is not good enough, in my judgment, to give an accurate internal surface replica.

Table 1 Volumetric Results

Specimen	Taxon	Cranial capacity (cm ³)	Method
OH 9	<i>Homo erectus</i>	1,067	Latex mould + plasticine
OH 12	<i>H. erectus</i> (?)	727	Plaster cast + plasticine
OH 13	<i>H. habilis</i>	650	Plaster cast + plasticine
OH 24	<i>H. habilis</i>	590	Plaster cast + plasticine + correction
KNM-ER 406	<i>Australopithecus robustus boisei</i> (male)	510	Volume formula + measurements
KNM-ER 732	<i>A. robustus boisei</i> (female)	506	Latex mould + plasticine

Average for *H. habilis*, including 650 cm³ value for OH 16, and 687 cm³ for OH 7, = 637 cm³.

(s.d. = 64 cm³, assuming coefficient of variation = 10%.)

Average for robust australopithecines, including OH 5 at 530 cm³ and SK 1585 at 530 = 517 cm³.

Average for gracile australopithecines¹¹ (n = 6) 442 cm³.

OH 9 and KNM-ER 732 were endocast directly from the originals using rubber latex; later small amounts of plasticine were added before determination of the total volume after first checking the dimensions of the endocast moulds against the originals. The differences in measurements were under 1%. In the case of OH 9, two reconstructions were made; the second followed Dr Alan Walker's skilful excavation of matrix from the middle and anterior cranial fossae. This permitted an unequivocal determination of the cranial volume. Only minimal modification was needed at the base and some plasticine was added to the parietal segments, particularly on the right side. Only the second reconstruction is given here. KNM-ER 732, comprising the right half of a presumably female robust australopithecine⁴, was endocast directly with latex stabilized with plaster; plasticine was added to form the missing occipital and cerebellar lobes. The small fragment of the occipital bone that shows part of the right occipital condyle, the position of the hypoglossal canal and an accessory vein, was utilized to reconstruct the region of the foramen magnum.

The volume of the KNM-ER 406 skull was estimated by using the formula⁵ $V = f \frac{1}{2}(LWH) + \frac{1}{2}(LWB)$, where f was obtained by using the average f s from OH 5 and from the new SK 1585 from Swartkrans^{6,7}. The dimensions of length (L), width (W), breadth (B) and height (H), were taken externally; estimates of bone thickness based on both KNM-ER 406 and OH 5 were subtracted from the external measurements. These dimensions should closely approximate the true internal endocast dimensions of KNM-ER 406.

Of all the reconstructions made, only OH 12 could show considerable error, because it has the most plasticine addition. This involves all of the frontal lobe and the temporal lobe. The frontal and temporal reconstructions were made by placing the right temporal fragment, showing part of the foramen ovale and the middle meningeal artery, in correct relation with the remaining main parietal fragment. The bregmatic and frontal portions were similarly placed in the

best position in relation to each other and the remaining endocranial reconstruction.

The results show (Table 1) that the volumes for the Olduvai hominids are either the same as or very near to, those given by Tobias^{2,3} whose estimates were made by the method of partial endocasts. Table 2 gives some of the statistical results using Student's t test for small samples⁸, using standard deviations based on the assumption of a coefficient of variation of 10%. Naturally the small sample sizes involved make this very speculative. Nevertheless, even with a small sample size and large standard deviations, the values of " t " are still very high. Assuming that the mean values are reasonably close to the true means, the values of " t " would become considerably higher as the sample size increased, and it is doubtful whether the standard deviations would go beyond a figure giving more than a 10% coefficient of variation. OH 7 and 16 have been included as part of the habilis sample, using the values given by Tobias^{2,3}. Although the value of 650 cm³ for OH 16 may be doubtful in view of the fragmentary nature of the skull, it is unlikely that it is grossly inaccurate if the large size of the frontal portions and the associated dentition are considered. The high value given for OH 7 (687 cm³) does not in any way seem extreme because no acceptable alternative ways of orienting the parietals could be found⁹. Furthermore, the parietals of OH 7 more than cover the completed OH 13 parietal regions on the reconstructed endocast (which gave a value of 650 cm³) and came closest to fitting the 727 cm³ reconstruction for OH 12 thus giving added support to the value found earlier by Tobias.

Table 2 Statistical Results

	Student t test (two tailed) ⁸		
	t	P	
(1) "Gracile" vs "robust" australopithecines*	2.46	<0.02	<0.05
(2) "Robust" australopithecines vs <i>H. habilis</i>	2.91	<0.02	<0.05
(3) "Gracile" australopithecines vs <i>H. habilis</i>	5.76	<0.001	
(4) "Gracile" and "robust" australopithecines vs <i>H. habilis</i>	5.38	<0.001	

* With $n=6$ for both groups, $t=3.56$ $P<0.01$ >0.0011

While the study of the morphological attributes of these endocasts is still in progress, the volumes and endocast shapes support the concept of a more advanced hominid population both distinct from *Australopithecus africanus*, *Australopithecus robustus* and *Homo erectus*, namely *Homo habilis*.

It would be unwise at this time to draw conclusions about the relationships between the fossils on the basis of endocast shapes until they have been more thoroughly studied. The KNM-ER 406 cranium is extremely similar to the cranial remains of OH 5; both gross observation and external measurements indicate that KNM-ER 406 is slightly smaller than OH 5. The appearance of the collateral vein in the occipital portion of KNM-ER 732, the overall shape and small size of the endocast, as well as the more gracile nature of the skull all suggest that KNM-ER 732 is most likely to be a female robust australopithecine skull¹⁰. The value of 506 cm³ for ER 732 represents 95% of the 530 cm³ values for OH 5 and for SK 1585, a value also consistent with sexual dimorphism as an explanation of the difference.

The low value of 732 cm³ for endocranial volume of OH 12 (VEK IV), obtained from such a high geological level (Bed IV) in relation to the 1,067 cm³ value for OH 9 (Upper Bed II), raises some interesting questions. Are these values simply the extremes of a range? Is OH 12 a remnant of the earlier *Homo habilis* taxon? Is it simply a small female *Homo erectus*? On the basis of the skull alone, OH 9 was certainly a large

and very robust hominid. The occipital and other skull fragments of OH 12 are very thick, and one fragment, that of the left petrous temporal, does not seem to belong with the other fragments attributed to OH 12, but must have come from a somewhat larger, second individual. Even accepting the OH 9 as a large male from the upper end of the range and OH 12 as a small female, there still remains a very considerable difference, and also a large hiatus between the Bed II habilines and OH 9. This, plus the announcement¹⁰ of a new skull, with an estimated cranial capacity of 800 cm³, dating from a 2.6 m.y. level from East Rudolf, would make any simple anagenetic explanation of hominids, existing as either single lines or types, as extremely speculative and unlikely.

I am indebted to the late Dr Louis, Dr Mary Leakey, Mr Richard Leakey, Dr John Harris and all of the staff at the Centre for Prehistory and Palaeontology in Nairobi, Kenya, for their kindness and cooperation, and to Dr Alan Walker for his help and advice in making these reconstructions. This work was supported by a grant from the National Science Foundation.

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Microbial CO₂ Fixation in the Anoxic Zone in a Volcanic Acid Lake

MEASUREMENT of CO₂ uptake using the ¹⁴C technique¹ in a volcanic acid lake Katanuma, in northwestern Japan, showed that microorganisms assimilate CO₂ in dark and anoxic conditions even though the pH of water is as low as 1.8.

Katanuma is a volcanic acid lake with an area of 0.1 km² and a maximum depth of 25 m. The pH of water ranges from 1.8 to 2.0, and the bottom is covered with sulphur precipitated from lake water by oxidation of H₂S. Volcanic gases, H₂S and CO₂, are supplied from fumaroles at the lake bottom. The lake water circulates from surface to bottom in spring and autumn, and stagnates in summer and winter when the lake is covered with ice. In summer (July 1972), the anoxic layer occurred below 4 m, and a slight amount of H₂S was detected in it but, in winter (January 1970), under ice cover, dissolved oxygen was found only down to a depth of 0.5 m, and an accumulation of H₂S was observed below 1 m depth. The electrode potential value calculated for the anoxic water of 20 m depth in winter was -27 MeV.

The CO₂ fixation was determined by the ¹⁴C technique¹, and CO₂ concentration in water was measured with an infrared CO₂ analyser². Results showed that in the dark, in summer, CO₂ fixation of the oxic layer was very low although the photosynthetic CO₂ fixation was rather high (8.92 mgCi m⁻² day⁻¹).

On the other hand, in the anoxic layer there was no indication of photosynthesis, but a considerable amount of CO₂ fixation in dark (19.08 mgCi m⁻² day⁻¹) was observed. In winter a very low photosynthetic activity was determined just below the ice, while a measurable amount of CO₂ fixation in dark (1.24 mgCi m⁻² day⁻¹) was obtained from the anoxic layer. The presence of microorganisms was observed microscopically in samples from anoxic layer, but taxonomic and physiological studies have not yet been completed.

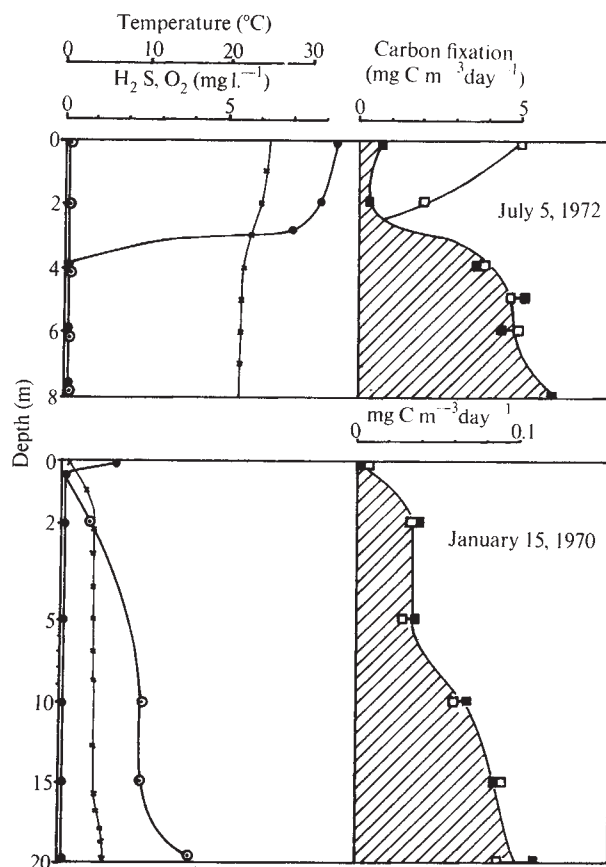


Fig. 1 Vertical distribution of carbon fixation (□, light bottle; ■, dark bottle), oxygen (●), hydrogen sulphide (○), and temperature (×) in Lake Katanuma.

So far as we know, there is no information about the existence of CO₂ assimilating microorganisms in such anoxic and strongly acidic water. Thiobacilli, iron bacteria and algae can survive in acidic water of about pH 2, but not in anoxic conditions. On the other hand, *Desulfovibrio* is active in an anoxic environment but only in water above pH 4 (refs. 3 and 4).

Our evidence of microbial CO₂ uptake in the dark, in anoxic and strongly acid water provide significant information regarding the limit of existence of microorganisms in the natural environment.

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Granules Containing Lead in Isolated Mitochondria

THE divalent cations of calcium, strontium, manganese, barium and magnesium can accumulate in mitochondria¹. In spite of variations in rates and in requirements for chemical components of the incubation medium, uptake of these cations leading to granule formation is dependent on energy¹. We report here the accumulation of lead in granules produced in isolated rat liver mitochondria following incubation in media containing lead. The computed levels of free lead ion in the media ranged from 0.2 μmol to 100 μmol ; by comparison, total plasma lead levels of up to 8 μmol have been found in victims of lead poisoning².

Mitochondria were isolated with as little disruption as possible by layering a 0.25 M sucrose homogenate on 0.34 M

0.1 mM; this was also possible using 0.2 mM lead nitrate and 0.3 mM aspartic acid instead of the previous lead-NTA mixture. Under these conditions significantly larger lead granules were formed (Fig. 1b).

For electron microscopy, samples were cooled to 4° C, spun at 2,000g for 5 min, washed with 0.25 M sucrose and centrifuged for a further 5 min at 2,000g. The pellets were resuspended, and fixed, in veronal acetate-buffered 1% OsO_4 for 0.5 h, dehydrated in acetone and embedded in 'Durcupan'. After sectioning on a 'Reichert' ultramicrotome the specimens were examined using a Philips EM 200 electron microscope.

The granules that were formed when the mitochondria were incubated in the media containing lead were extremely dense and could be seen in the matrix around the cristae and other regions of the inner membrane. Incubation of mitochondria in the presence of 4 mM CaCl_2 led to the formation of granules that were less electron-dense (Fig. 1c), whereas if lead and calcium were both absent no granule formation was observed. Other investigators^{1,4,5} have shown that calcium ions are actively accumulated by the mitochondria incubated under similar conditions and that the resulting electron-dense granules have a tertiary calcium phosphate composition. Other calcium phosphate-containing mitochondrial granules have been observed in many cell types—including chondrocytes—under normal physiological conditions^{6,7}. It has been suggested that the potential for calcium uptake by mitochondria is ubiquitous in the tissues of higher animals⁷.



Fig. 1 Electron-dense granules (arrows) in isolated rat liver mitochondria. Osmium-fixed, unstained. a, Granules containing lead produced with NTA-metal buffer. $\times 62,000$. b, Granules containing lead produced with aspartic acid-metal buffer. $\times 62,000$. c, Granules containing calcium. $\times 69,500$.

sucrose and centrifuging at 500g for 10 min in a Sorvall RC-2 refrigerated centrifuge. The supernatant was then spun at 2,000g for 10 min to recover the mitochondrial pellet which was used in these experiments. As controls, mitochondria were incubated at 37° C for 30 min in the following calcium-free medium: 80 mmol NaCl, 10 mmol MgCl_2 , 10 mmol succinate and 5 mmol ATP adjusted to pH 7.2 with 10 mmol Tris-HCl and 4 mmol phosphate buffers. The experimental samples were incubated in this medium with the addition of 1 mmol lead nitrate and 1.5 mM nitrilotriacetic acid (NTA). The granules formed under these conditions are illustrated in Fig. 1a. (The concentrations of lead and NTA were chosen so as to avoid precipitation of lead chloride and lead phosphate in the medium.) Using published pK and stability constant data for lead ion with the ligand NTA³, the amount of free lead ion in the solution was calculated to be 2×10^{-4} mM at pH 7.2. If the phosphate was omitted from the medium and the NaCl replaced with an equiosmolar concentration of sucrose, the level of free lead ion could be raised to a computed level of

When succinate and ATP were omitted from the incubation media containing lead, lead was not observed within the mitochondria. Addition of antimycin A, dinitrophenol (DNP), or sodium azide to the media at the beginning of the incubation period resulted in significantly fewer and smaller dense granules. These substances are metabolic inhibitors which operate at different points in energy pathways. Ouabain, which is an inhibitor of some K^+ -dependent ATPases but which has little effect on mitochondrial breakdown of ATP, was inactive. These results imply that the mitochondrial uptake of lead is a process dependent on energy.

A difference between calcium and lead-rich granules was observed in further incubation for 60 min in the basic medium containing 0.5 mM DNP. Whereas calcium granules became largely dispersed⁵, the lead granules seemed to be unaffected, indicating that lead removal is a less readily reversible process. This is consistent with the observation that lead binds much more strongly than calcium to some ligands (for example, sulphhydryl and phosphate groups³). It would be expected

therefore that if mitochondria accumulate lead in clinical lead poisoning as they do calcium under normal conditions, the subsequent removal of lead from the mitochondria would be physiologically difficult.

It is suggested that the effects of lead on the mitochondria are two-fold. First, by analogy with calcium uptake¹, diversion of energy derived from the electron-transport chain to the active accumulation of lead would be expected to prevent ATP synthesis and to interfere with the maintenance of ionic concentration gradients across the membrane barriers⁸. Second, lead in the mitochondria would be expected to hydrolyse ATP⁹, form complexes with the SH⁻ groups of mitochondrial enzymes, such as those involved in cytochrome synthesis, and to react with other matrical anions including phosphates. The terminal result of excessive accumulation of lead cations by an intact cell would thus lead to a disruption of the ability of the cell to maintain itself metabolically and structurally. This failure would result in death and dissolution of the cell. These processes may account for the fibrosis observed in the liver and kidney of lead-poisoned victims.

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GENERAL

New Problem in Concept Formation

THERE is a simple rule that defines which of the symbols of Fig. 1 fall into group A and which into group B. It can be expressed in thirteen words, but the reader is unlikely to guess it—even if we tell him that the only relevant differences between the patterns are in symmetry, dot colour and dot shape.

The rule is that an A symbol is at least two of symmetric, black and composed of circles. The difficulty is that no single property is necessary or sufficient to make a symbol a member of either group. We therefore call this a polymorphous rule, following Ryle¹ and Urmson². We can define an *m*-out-of-*n* polymorphous rule as one in which there are *n* relevant conditions of which *m* must be satisfied; Fig. 1 involves a two-out-of-three polymorphous.

We have done several experiments to prove that polymorphous rules are exceptionally difficult to find. We report two experiments here: they compare the polymorphous rule with the disjunctive and conjunctive rules studied by Bruner, Goodnow and Austin³. A conjunctive rule (for example "an A is both black and symmetric") has necessary but no sufficient conditions; it could be called an *n*-out-of-*n* polymorphous. A disjunctive rule (for example "an A is either black or symmetric") has sufficient but no necessary conditions; it is a one-out-of-*n* polymorphous.

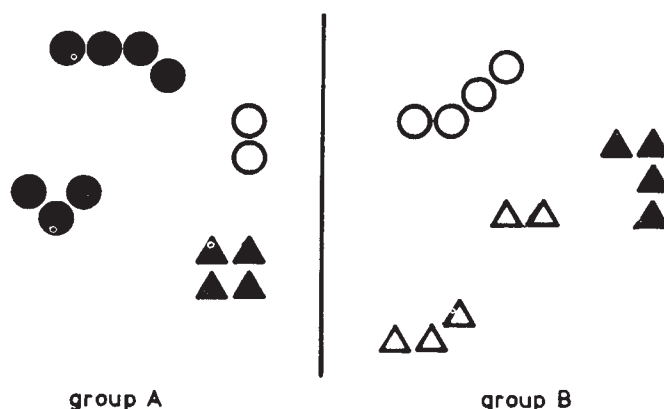


Fig. 1 Patterns determined by a two-out-of-three polymorphous rule.

In experiment 1, seven undergraduates each tried to solve three problems. In each, they were given a pack of forty-eight cards and told to expose them one by one. They were told that the pattern on each card belonged to one of two groups, A and B, and that they must try to find and use the rule that determined which patterns belonged to group A. As the subject exposed each card, he guessed whether it belonged to group A, and was told "yes" or "no". If he made eight consecutive correct guesses he was asked to state the rule as best he could. Any rule was accepted if it correctly assigned all forty-eight cards without simply enumerating them; giving an acceptable rule ended the test.

Table 1 Dimensions of Variation in the Three Packs of Stimulus Cards

	Pack 1	Pack 2	Pack 3
Nature of material	Arrays of type-written letters	Rows of shapes	Random shapes
Dimensions	Colour (black/red)	Number in row (odd/even)	Outline (smooth/jagged)
	Symmetry of array (symmetric/not)	Shape (square/circle)	Colour (blue/green)
	Letter (x/o)	Shading (filled/empty)	Shading (hatched/empty)

Each subject faced one conjunctive, one disjunctive and one two-out-of-three polymorphous problem. In each there were three two-valued dimensions of variation in the patterns, but in the conjunctive and disjunctive problems one of these was irrelevant. The experimenter told the subject what the three dimensions were. Three packs of cards, using different sets of dimensions were used; the symbols are summarized in Table 1. All six possible orders of presentation of the three problem types were used, and the assignment of card pack to problem type was balanced between subjects. For all problems, there were twenty-four group A cards in the pack.

All seven subjects performed worst on the polymorphous problem. The median numbers of cards exposed before the last incorrect guess were nine, twenty-eight and forty for conjunctive, disjunctive and polymorphous problems respectively. Four subjects did not reach the criterion on solution on the polymorphous problem before the pack was exhausted; those who did solve it gave the rule as a disjunction of conjunctions (for example "an A is black and composed of circles, or symmetric and composed of circles, or black and symmetric") rather than in the simpler "two-out-of-three" style.

The concordance between subjects as to the order of difficulty of the problem types was 0.8 (Kendall's W coefficient of ranks⁴), significant at the 0.001 level. There was no

effect of order or pack, and other measures of difficulty lead to similar concordances. Experiment 1 thus showed that disjunctive problems are harder than conjunctive ones (confirming a result of Bruner *et al.*), and that polymorphous problems are harder than either.

In experiment 1, subjects could only acquire information gradually. In experiment 2, all possible information was available from the start. Six more undergraduates served. Cards were selected from the packs used in experiment 1 to make packs of eight including every combination of the three stimulus dimensions. These were laid out on a table, segregated into groups labelled A and B. The experimenter told the subject that a simple rule determined whether a card belonged to group A, asked him to find it as quickly as possible, and measured the time until he offered an acceptable rule. Unacceptable rules were refuted by pointing to counter-examples. The maximum time allowed was 10 min. Each subject faced one problem of each type (conjunctive, disjunctive and two-out-of-three polymorphous); order and pack were again counter-balanced, and the subjects were told before they started each problem what three dimensions might be relevant to it. Their task was thus like the puzzle we set the reader at the beginning of this communication.

Once again, every subject did worst on the polymorphous problem. The median times to solution (scoring 10 min for complete failure) were 34 s, 2 min 35 s and 10 min for conjunctive, disjunctive and polymorphous problems respectively; four subjects failed to solve the polymorphous problem (the other two gave the rule as a disjunction of conjunctions). Kendall concordance between subjects as to order of difficulty was 0.63, significant at the 0.05 level⁴. There was no effect of pack or order of presentation. The two experiments lead to the same conclusion—that polymorphous rules are very hard to discover. The total failures to solve the polymorphous problems prevent us setting figures on their difficulty, but the direction of the result is unambiguous.

Linguistic philosophy gives peculiar importance to the polymorphous rule. Ryle^{1,5} suggests that many philosophically important concepts (such as "thinking") and everyday terms (such as "solicitor") have polymorphous definitions, whereas Wittgenstein⁶ seems to suggest that such definitions are the rule rather than the exception. Also Neisser⁷ has remarked that written letters and spoken syllables, the patterns we recognize most easily and commonly, fall into "ill-defined categories". It is notable that all these polymorphous concepts are generated by human behaviour; and in experiments on spontaneous classification, we have found that subjects use polymorphous rules quite freely. It seems extraordinary that such rules should be so hard to formulate. Yet there is no reason to think the difficulty is not general. We report here experiments using two different test procedures and three different sets of stimulus material; informal tests under the most varied conditions have always produced the same result. Nor is the difficulty entirely in the formulation of the rule: the trials-to-last-error measure used in experiment 1 does not involve explicit formulation. At the least, these experiments point to a difference between rules which are easy to use and those which are easily learned; it may be that this reflects a difference between rules which we generate spontaneously and those which we can extract easily from external problems. This would be evidence against the widely held theory of "analysis by synthesis" (see Neisser⁷, page 161ff), which implies that rule discovery inevitably involves internal generation of a corresponding rule.

We thank Dr Hall for discussion.

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Comment

THE preceding communication presents two experiments which investigate the ability of undergraduates to deduce and formulate a classification rule for three problem types. The performance of the subjects is described in terms of two quantitative measures—median number of cards exposed in experiment 1 and median time to solution in experiment 2. As the authors admit, however, the actual numerical values presented are rather misleading because subjects fail to achieve a solution. I cannot understand why the authors did not present detailed qualitative descriptions of the kinds of rules offered by the subjects in each task.

The chief conclusion of the communication is that in the experimental conditions used by Lea *et al.*, *m*-out-of-*n* polymorphous problems are very difficult to formulate. The authors remark, however, that some workers have claimed that polymorphous rules are those most frequently used in normal cognitive activity and they mention that they have obtained experimental evidence (in conditions which are not clearly specified) which suggest that subjects can and indeed do choose to use polymorphous rules frequently. It seems to me that the authors should ask themselves why, with their experimental procedure, instructions, and stimulus material, the subjects failed to show the supposed fluency with polymorphous problems. It is worth noting that several workers have remarked that we habitually categorize and assign structure to events using rules which we cannot formally define—see work on visual and auditory pattern recognition and interpretation, and linguistics.

Lea *et al.* argue that because they obtained their results using two "different" test procedures (by in fact varying the number of visible stimuli against which the subject could evaluate a rule), three different sets of stimuli (by varying the selection of relevant stimulus dimensions but not by sampling different classes of stimuli) and in informal tests under the "most varied conditions" (unspecified) there is no reason to doubt the generality of the finding. This statement may or may not be correct but the authors give little information with which to evaluate their claim. The authors finally suggest that the apparent conflict between their results and the supposed generality of polymorphous concepts may reflect the fact that polymorphous concepts are hard to learn but, once learned, are easy to use. The discussion which follows this remark contains a number of implicit assumptions which to my eyes move the discussion into the realms of the inscrutable.

The interest of the authors' claim must depend on how adequately an experimental procedure which involves the presentation of meaningless patterns yields a representative measure of the human capacity to deduce and test a conceptual role.

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BOOK REVIEWS

History of Biochemistry

Molecules and Life: Historical Essays on the Interplay of Chemistry and Biology. By J. S. Fruton. Pp. x+579. (Wiley-Interscience: New York and London, November 1972.) £8.40.

FRUTON gained general respect because of his early work on peptides which led to the synthesis of substrates for various proteolytic enzymes—including pepsin. A generation of students is indebted to him, and Simmonds, for a text book of biochemistry. Now he has written an excellent history of biochemistry. Even finding out the dates and first names of nearly 1,500 people listed in the index must have been a formidable labour. But the book is not just a catalogue of scientists and their work, it is a closely argued discussion of their philosophical outlook, influence and obsessions. On the whole, Fruton is kind. He concentrates on the discoveries that scientists made rather than on their mistakes and illusions. He says that he has tried to avoid classifications into "good guys" and "bad guys" and apologises for any lapses into the "Whig interpretation of history" which sees developments, not through contemporary eyes, but by hindsight. So as to avoid too much influence of hindsight, the copious quotations he includes, dealing more with research than with general survey, are usually taken from early papers rather than from autobiographies or accounts written long after the research was completed. Fruton mentions several examples of faulty memory, or the wish to present a tidy account, leading scientists, writing late in life, seriously to misstate the actual steps by which they made discoveries.

The book does not consider the whole of biochemistry in a continuous historical narrative. Instead it is divided into six sections. The first deals with the physical environment in which biochemists worked. Fruton traces the development in different countries of research by amateurs, in universities and in research institutes; the development of scientific societies; and the proliferation of journals.

The study of fermentation may be

regarded as the beginning of biochemistry: the importance of yeast in our subject is therefore fittingly commemorated in the derivation of the word enzyme. Enzymes are the subject of Fruton's second section. He traces the slow acceptance of the idea that fermentation was one of yeast's ways of life, the establishment of the idea as a dogma, and the demolition of the dogma when Buchner, who was trying to make an antitoxin, got fermentation with a cell-free yeast extract. In Jacques Loeb's phrase, "Biology was relieved of another fragment of mysticism."

Emil Fischer gets most citations in the index, perhaps because Fruton's main personal interest is peptides. Liebig comes second. He dominated the mid-19th century, both because of his sound work on pure chemistry and on enzymes, and because of his forcefully expressed but often erroneous opinions on agriculture and metabolism. He did not escape criticism; thus Berzelius wrote: "This easy kind of physiological chemistry is created at the writing desk, and is the more dangerous, the more genius goes into its execution . . ."; Lawes Gilbert demonstrated the falsity of his views on fertilizers and on protein as the source of muscular energy. It is nevertheless clear from Fruton's account that Liebig's influence was, on the whole, beneficial. This leads to the depressing conclusion that there is validity in Francis Bacon's aphorism, "Truth often comes more easily from error than from confusion."

Those who yearn for scientific probity will get more comfort from the third section—on proteins. Eminent scientists are shown persistently to have enmeshed themselves in false assumptions without deluding the mass of their contemporaries. Willstatter thought that enzyme activity was caused by a little something added to a carrier, Bergmann thought that the sequence of amino-acids in a protein must be regular, Langmuir thought that the final configuration must have symmetry and regularity. Other physicists are quoted who, having donned a biological strait-waistcoat

of their own invention, grumble because it is uncomfortable. One of the few criticisms I have of this book is that Fruton, when discussing the crystallization of enzymes, does not comment on the curious absence of tests for activities, other than that sought, in the early crystalline preparations. Some carried so many enzyme activities that it was simply preconception that gave them one name rather than another.

The effects of erroneous assumption and premature generalization are important features of the section on nucleic acids. As with proteins, there is the intrusion of the idea that they must be simple and regular. Liebig was exceptional in thinking that proteins from different organisms were often identical, but many biochemists assumed that there were just two nucleic acids, and that all plants contained one and all animals the other. Fruton points out that towards the end of the 19th century nucleic acids were assigned extensive roles, it was false assumption that led to their neglect during the first half of this century.

The fifth section traces the slow development of clear ideas about biological oxidations. Progress here seems to have been inhibited much more by the intrinsic difficulty of doing relevant experiments with biological material than by false assumption. Even so, it is interesting that as late as 1914, W. B. Hardy felt constrained to write, "It must always be remembered that an oxidation place is also a reduction place, and it is to be called the one or the other according to the particular zero which is chosen." Considering the part that spectroscopic measurements now play in the study of enzyme actions and in the identification of substances, it is salutary to remember that MacMunn stressed in 1886 the usefulness of coloured substances in studying the internal processes of tissues: the technique was not extensively used till after Keilin's publications from 1925 onwards.

Some aspects of metabolism and of the internal organization of the cell are

the themes of the last section. At one time hypothetical entities, of which protoplasm was the most respectable, were invoked to "explain" everything. Then Hopkins led the revolt, pointed out that these entities were merely words and explained nothing, and called for comprehensible reactions between small molecules. Hopkins may have overreacted against mere size in molecules – but he got rid of a great deal of notional biochemistry and of the idea that biology involved peculiar processes that could not ultimately form part of sciences such as chemistry and physics. Among the many quotations in this excellent book is one from Chevreul in 1824, "Are the nature and effects of physical and chemical forces sufficiently well known at present to allow us reasonably to affirm that a particular phenomenon of living bodies is absolutely independent of them? Certainly not." That is still the basic postulate of biochemistry.

N. W. PIRIE

Roots of Astronomy

Education in and History of Modern Astronomy. Edited by R. Berendzen. Pp. 275. (New York Academy of Sciences: New York, August 1972.) \$24.

AT the end of the eighteenth century, an observer of the educational scene in Britain remarked, "It has long been a matter of surprise to those who are interested in the education of youth, that . . . so few attempts have been made to facilitate the study of astronomy". The appearance of the proceedings of this first international conference on education and history of modern astronomy allows us to ask the question—how do things stand now?

Although the present volume does report briefly on astronomical education in the United Kingdom (and also contains statements of the position in Australia, Canada and India), the main emphasis is, hardly surprisingly, mainly on astronomical education in the United States. One gains the overall impression that, though the amount of astronomy teaching at all levels in the USA may not be very large, much more effort is being expended on its development than in the UK. Thus the American work on astronomical films, CCTV, computer graphics, case-study projects and the educational use of planetaria finds as yet only few parallels in Britain. On the other hand, postgraduate astronomical education in the United States seems to face more problems than ours does, since both the relative expansion in postgraduate numbers has been greater there, and the cutback in funding has proved relatively

more severe. Astronomy students in both countries might, perhaps, envy recruits to traditional astronomy courses in India who, we are told, can usually find employment "as teachers or start their own businesses as astrologers and/or priests". (On the debit side, these Indian courses have the disadvantage of being taught only in Sanskrit.)

One of the most important emphases in US astronomical teaching lies in the attempt to involve students who have no intention of specializing in astronomy: indeed, who have no wish even to specialize in science. A good deal of imagination is evident in the handling of such students. Thus the University of Southern California runs a course entitled "Astronomy's Contribution to Western Culture". The various projects thought up by students include one—described as "most stimulating"—consisting of a study "that one of our teetotaling students performed on the effect of brandy on limiting magnitude". There is clearly a fairly widespread belief amongst US astronomers that introductory courses of this type, apart from their intrinsic worth, help counter the growing anti-scientific trend amongst American students, not least as regards the revival of astrology. (One of the reported discussions at this conference provided, incidentally, a fine definition—culled from a woman's magazine—of the difference between astronomy and astrology. "Astronomy is when you go outside and look at the sky, and all you see are the stars. Astrology is when you go outside and you look at the sky, and you see dragons and virgins and other creepy things.")

It is evident from the papers and discussions at this conference that putting on broadly-based, culturally relevant courses actually entails more work than putting on specialized astronomy courses. One main reason is that an astronomer's research interests are not normally relevant to the former type of course. Nevertheless, many of the American participants obviously thought the effort worthwhile. What then, in comparison, of the British scene? It seems to me that we fall down just at this point where most emphasis is being brought to bear in the United States. Our education of would-be specialist astronomers is reasonably good, but our provision of astronomy as a non-specialist subject, and at levels below the university, still leaves a great deal to be desired. Surely, interest in the physical sciences is not so vast that we can afford to ignore astronomy, currently one of the most fascinating sciences for the layman? The effort involved in a really major increase of astronomical teaching at an introductory level would certainly be large, but it might prove to be eminently worth making.

A. J. MEADOWS

Electrodes

Electrochemistry. Edited by J. O'M. Bockris. Consultant Editor A. D. Buckingham. Pp. 332. (MTP International Review of Science.) (Butterworth: London, University Park: Baltimore, 1973.) £10; \$24.50.

A NOTE by the publishers of the *MTP International Review of Science* states that the aim is to provide regular authoritative reviews of entire disciplines. This volume is the first on electrochemistry and it is devoted almost exclusively to what is sometimes termed electrodes: the study of phenomena and reactions at electrode surfaces. Within this narrow field, which very much mirrors the interests of the editor of the present volume, some useful contributions have been brought together. The first chapter deals with the fundamental theoretical problem of electrodes—the mechanism of the transfer of charge between ions and an electrode surface—and it traces the development of ideas since the first introduction of a quantum mechanical model by Gurney in 1931. This is very much a chapter for specialists but it is useful to have ideas which are otherwise scattered through the literature brought together in a systematic survey. A disappointment is the cursory dismissal of modifications of earlier ideas which the discovery of the hydrated electron may well bring by the *ex cathedra* statement of the authors that in their view "the solvated electron model is to be rejected for normal electrode processes". In the second chapter Professor Conway considers the mechanism of electrode reactions in a more experimental context and particularly where adsorbed species play an important role; this too is useful, but it is inevitably an up-dated and condensed version of his well-known book on *Electrode Processes*. The survey then changes direction towards metallurgy, and the effect of electrochemical factors on the strength and stability of metals and the participation of metal surfaces in dissolution and deposition processes are considered. An interesting account of hydrogen in metals follows, and this is succeeded by a very detailed survey of the investigation of metal surfaces, particularly at electrodes, by ellipsometric optics. This is highly mathematical, and not easy reading, but it provides a mass of useful material. The final chapter, with an abrupt change of theme, takes an all-round look at electrochemical participation in biological processes; it gives a good insight into what may well prove to be a fruitful electrochemical field.

To sum up, what the survey does is good and it provides rich food for specialists on a miscellany of topics mainly related to electrochemical processes at surfaces, but it is certainly not

a systematic survey of the present state of electrochemistry as a whole, and a reader who refers to it with this hope will be disappointed. Printing and illustration are excellent, but there is no index.

A. HICKLING

Environment of Leicester

Leicester and its Region. Edited by N. Pye. Pp. xxi+603+21 plates. (Leicester University: Leicester, September 1972.) (Published for the Local Committee of the British Association.) £6.

FOR many years the annual meetings of the British Association for the Advancement of Science have been marked by the publication of a special volume dealing in a scholarly way with many facets of the host city and its surrounding region. This has usually been promoted by a special local committee in which "town" and "gown" have collaborated in an exemplary fashion, and the general editor has usually been the University's Professor of Geography. The Leicester meeting of 1972 continued these traditions in a most impressive way by the publication of a superb volume containing 600 pages, with ninety-three figures, forty-eight tables and over twenty plates, which, in both size and scholarship, aptly matches the large volume on Nottingham and its region produced for the annual meeting of the Association in 1966. Thus, after a 6-year wait, Leicester has drawn even with its traditional East Midland rival.

It is difficult, of course, if not impossible, to review a work of such length and variety in a conventional way. One can only say that Professor Pye, the editor, and his thirty-five contributors, drawn predominantly from the academic community, but also including representatives of local government and other walks of life, have fulfilled their task with great credit. Each chapter is an up-to-date, authoritative statement on a particular aspect of the city and county that in most cases draws skilfully on the published literature while at the same time offering an additional contribution to our knowledge, whether through fact or commentary.

The book falls into several main sections. After an introduction by Professor Pye, part 1 deals with the physical background (geology; geomorphology; weather and climate; soils; flora and vegetation; fauna; and nature conservation), and part 2 traces the area's historical development (prehistoric and Roman settlement; Saxon and Danish Leicestershire; Leicestershire 1100–1800; the evolution of Leicester 1066–1835; and Leicester and Leicestershire 1835–1971). Part 3 is devoted to economic structure (communications and transport; agriculture; extractive

industries; foundation of the modern industrial pattern; the modern industrial scene; science, technology and local industries; patterns of population change; and urban structure of Leicester). Finally, part 4 deals with social development (schools in Leicester; education in Leicestershire; further and higher education; the development of medicine and social services; the arts; vernacular architecture; and the planning of the Leicester of the future).

While the volume is intended to offer a study of a particular English industrial city and its hinterland, the specific experience and characteristics of Leicester and Leicestershire nevertheless almost constitute a "model" case. Here in the very heart of England we have a county still firmly dominated by its ancient capital, which is surrounded by a number of subordinate urban centres with their own local spheres of influence—the epitome of a geometrically-arranged hierarchy of "central places". The physical environment includes a representative sample from the geological time scale, including both the Pre-Cambrian and the Pleistocene. There is a fairly wide range of mineral resources, including coal, ironstone, roadstone and brick earths. Leicester itself exhibits tangible evidence of all the major phases of English urban development, including the remains of the Roman town of *Ratae Coritanorum*, mediaeval churches and Guildhall, Georgian town houses, Victorian working-class factory districts, and the suburban sprawl of the present century. (It is scarcely surprising that the local College of Education has sponsored the idea of "town trails" in Leicester.) Modern Leicester is large enough to exhibit the well-differentiated commercial, industrial and residential neighbourhoods so typical of Western cities (and thus incidentally lends itself to factor analysis and other techniques of the modern urban analyst). The city's industrial evolution shows the typical change from a varied structure of numerous small handicrafts and trades to reliance on one or two dominant manufacturing industries, in this case hosiery and footwear, that began as domestic activities and became converted to a steam-powered factory basis after the middle of the nineteenth century. The twentieth century has dealt kindly with Leicester, so that, unlike the experience of some northern industrial towns, the spontaneous growth of new industries, dominated by engineering, has diversified a potentially vulnerable industrial structure and taken up the slack caused by the falling or static labour requirements of hosiery and footwear.

While there is so much that is typical of the country as a whole, there are,

however, important achievements and innovations to Leicestershire's own credit. For example, modern livestock breeding was pioneered by Robert Bakewell. The hiring of a special train from Leicester to Loughborough by Thomas Cook marked the advent of the modern tourist movement. Frank Whittle nurtured his jet engine in local factories during the late 1930s and early 1940s. And, quite understandably, important developments took place here in the technology of hosiery and footwear manufacture. More recently, the "Leicestershire Plan" for education, leading to the establishment of separate upper schools for all pupils over 14, and the "Leicester Traffic Plan", involving the segregation of pedestrians and vehicles and the shielding of the city centre from the motor car, have both achieved fame for their bold imagination.

Leicester and its Region will be used chiefly as a valuable reference book for the upper forms of schools and for institutions of higher education, especially where geography, local history and environmental studies are concerned. In that role it will, over many years to come, handsomely vindicate the labours of all those who have contributed to it.

R. H. OSBORNE

Catecholamines

Handbook of Experimental Pharmacology/Handbuch der Experimentellen Pharmakologie. Edited by H. Blaschko and E. Muscholl. Pp. 1,054. (Springer: New York and Berlin, 1972.) DM396; \$125.60.

THIS tome on the catecholamines is volume 33 of the new version of the famous Heffter's *Handbook of Experimental Pharmacology*. The volume has 1,054 pages and its weight makes "Handbook" a curious misnomer. But the weight is justified by the contents. The enormous increase in catecholamine research represented here is due not only to the universality of their distribution and effects but also because the analysis of their mode of action led to the discovery of the cyclic AMP system which is now recognized as a common mediator of the actions of many other hormones as well. Appropriately, therefore, a masterly account of the effects of catecholamines on metabolism is given in a central chapter by Jean Himms-Hagen, much of which summarizes present knowledge on the effects of catecholamines on adenylyl cyclases and on enzymic effects of cyclic AMP.

All chapters are of high quality in both content and style, as would be expected from the well-selected experts who wrote them. It would be invidious to select any particular chapter for praise, but to someone working on

amine storage and release mechanisms, the two chapters devoted to these could hardly be improved on.

This book should be available to everyone engaged on any aspect of catecholamine research. The pages are packed with essential information which is remarkably up to date for such a compendium; apparently most contributors were able to include consideration of publications in the first half of 1970. Another attractive feature, traditional for this handbook, is the freedom with which the contributors lay bare their ideas on mechanisms and provide stimulating but concise expressions of opinions about open problems. Their involvement in current research makes these opinions very much worth having. The editors, H. Blaschko and E. Muscholl, deserve congratulations and gratitude from all those interested in the catecholamines. The publishers deserve congratulations for the excellence of production of the book which includes a detailed, accurate index.

G. V. R. BORN

Separation Techniques

Pharmaceutical Applications of Thin-layer and Paper Chromatography. Edited by Karel Macek. Pp. xvi+743. (Elsevier: Amsterdam, London and New York, 1972.) Dfl. 250; \$73.50.

CHROMATOGRAPHY is the name given to a series of closely related but technically different separation techniques whereby minute quantities of mixtures can be resolved into their various components. The simplest and cheapest techniques—and thus the most popular—are paper and thin-layer chromatography. One consequence of the popularity has been the extraordinary number of publications dealing with one aspect or another of the technique or of application of the technique in the solution of different analytical problems. It is hard to find a branch of science that has not benefited from chromatography, although it could easily be argued that the pharmaceutical sciences have benefited most. A variety of chemical substances, synthetic and natural, will be found in the pharmaceutical literature: psychotropic drugs, antihistamines, antipyretics, analgesics, antitussives, sedatives, hypnotics, anaesthetics, anti-epileptics, local anaesthetics, adrenergic and cholinergic substances, diuretics and cytostatics to name but a few of the synthetic substances; alkaloids, glycosides, antibiotics, laxatives, vitamins, to name but a few of the natural products as well as auxiliary substances such as antioxidants, colouring matters and preservatives. The analyst may have to examine any one of these drugs for impurities or determine the amount of one in the presence of others. Thin-

layer and paper chromatography has almost certainly been used by someone and reported in the literature. The editor, who is bibliography editor of the *Journal of Chromatography*, in association with expert workers with the different groups of substances has compiled a very comprehensive review of more than 2,500 references. Each chapter begins with generalized information about extraction procedures for pharmaceutical dosage forms or from crude drugs and includes useful comments and explanations in connexion with the methods described. The free use of tables summarizes important information such as solvent systems, spray reagents and *R_F* values.

Prior to this main section of the book there is a short section on techniques—qualitative and quantitative, which, without resource to repetition of much information already detailed in other books, gives facts and advice that are equally useful to the expert and the beginner. The text flows remarkably well and it is only the heading of chapter 9—"The Tasks of Paper and Thin-layer Chromatography" that betrays its Czechoslovakian origin. It is an extremely good translation and those concerned are to be congratulated. The book is well bound, the text is clear and printed on good quality paper with few typographical errors. While it is doubtful whether many British analysts can afford to purchase it, there is no doubt that the book should be in every laboratory concerned with pharmaceutical analysis.

E. J. SHELLARD

Stress on Plants

Responses of Plants to Environmental Stresses. By J. Levitt. Pp. xii+697. (Academic: New York and London, March 1972.) \$32.50.

JACOB LEVITT is one of the leading scientists in the field of stress physiology of plants. As a sequel to his comprehensive studies on temperature and drought resistance of plants, Levitt presents a fifth far more detailed and thematically enlarged monograph, which covers most of the recent literature from all parts of the world up to 1969-70. The references run to nearly 100 pages.

It was not, however, the author's intention to write a compendium complete in every respect, but to give an account of the research according to his own particular point of view. This holds particularly for the sections concerning temperature and water stress, which takes up four fifths of the book. The remaining chapters deal with the influences of the different kinds of radiation, salts, and mechanical factors as well as stresses caused by human civilization.

In an introductory chapter Levitt

points out ways to a unified stress concept. To conform with interdisciplinary usage he has used a uniform terminology as, for example, the terms "tolerance" instead of "hardiness" or "freezing" instead of "frost". Levitt contrives to formulate the term "stress" which has its exact definition in physics, which is otherwise used in a less precise way. He makes a clear distinction between "stress" as an ecological agent and "strain" which implies the impact and the effect on the plant.

There are strains of an elastic nature, which occur repeatedly without, however, exhausting the plant, so that the effect is reversible. Or they may be of plastic nature, stimulating the plants into adaptation to the stress. Levitt deals mainly with the plastic strain. Resistance of plants can be brought about by avoidance or by tolerance of a strain.

This concept is applied to the different kinds of stresses set out in analogous schemes. In a concluding chapter Levitt discusses the problem of comparability of the strains of the different stresses. Desiccation, for example, can be caused by single or a complex of quite different stresses such as low temperature, retarded water conductivity, osmotic effects, heat, and so on. Levitt distinguishes between comparative primary, secondary and tertiary stresses. The capability of plants to avoid strains can be ascribed to different causes. But as to how plants tolerate the different strains certain correlations can be stated. Most of the stresses described above claim the tolerance of the plants in a similar way and will finally lead to the same injuries. Thus a uniform scheme (system) of injury could be considered possible. In this context Levitt offers his not entirely undisputed theory, developed in 1962, that those injuries are results of oxidative sulphur, sulphur bindings in particular at lipids and proteins—this is a new aspect—at the membranes, which lose their semipermeability. As discussed extensively in the chapter "Molecular basis of freezing injury and tolerance" this concept turns out to be consistent with the hardening of many plant species, which keep a high reducing capacity (that is, many sulphhydryl-groups). Levitt endeavours to integrate into his concept many other existing theories, or to refute them. The seasonal "phenological" view of hardening is relatively neglected in favour of the biochemical aspects.

This excellently written book will be an indispensable source of knowledge and information for physiologists, ecologists and all students of applied biology. It should also be considered as an impulse to further critical discussions about causes of the resistance of plants (and animals) to extreme environmental stresses.

L. KAPPEN

Obituary

Dr L. S. B. Leakey

LOUIS SEYMOUR BAZETT LEAKEY, M.A., PhD, DSc, FGS, FBA, died in London on October 1, 1972, while on his way to America to fulfil lecturing engagements. Perhaps best known as a physical anthropologist, palaeontologist and prehistoric archaeologist of distinction, he was also a man of many talents.

Louis Leakey was born in 1903, the elder son of missionary parents who were working among the Kikuyu at Kabete near Nairobi. From his earliest days his schoolmates were from this tribe so that his command of the Kikuyu language was unrivalled by any other European. Taught at first by his parents and a governess, Leakey was prevented by the Great War from attending an English school until he was over 16. In 1922, he entered St John's College, Cambridge, as a Sizar; some 44 years later he was made an Honorary Fellow of the College in recognition of his worldwide achievements. At Cambridge he studied Modern French, Medieval French and Kikuyu for Part I of the Tripos, an unheard of combination. He went on to take a first and then another in Archaeology and Anthropology. As a result of his academic performance at Cambridge and his experience in Africa he was awarded a research studentship that took him on a number of archaeological expeditions. Later, in 1929, he was made a Research Fellow of St John's and took his doctorate.

While at Cambridge he suffered a rugby injury and was advised to recover in the "fresh air". He took a year's leave and accompanied Cutler, a Canadian palaeontologist, who was just leaving for Tanzania to collect fossils for the British Museum (Natural History). This invaluable period in the field, under expert guidance, gave him both practical experience and a basic knowledge of palaeontology. At first Leakey's principal interest was in the sequence of prehistoric cultures in Kenya. He attempted to correlate cultural changes with climatic changes during the African Pleistocene in terms of pluvials and interpluvials corresponding with European glacial alternations. During this fieldwork several important, if controversial, finds were made, in particular the Kanam jaw and the Kanjera skulls. In each case the

antiquity of the material was doubted, and in one case even the provenance; this pattern of events is well known to human palaeontologists and to others in science whose discoveries disturb the tranquillity of textbook writers.

In the late 1930s Louis Leakey spent over two years in the study of the history and customs of the Kikuyu tribe. The results of this extensive research were written many years ago and they will be published soon as one of his posthumous papers. After the 1939-45 war, in which he served in the African section of the Special Branch CID, Leakey was appointed full-time Curator of the then Coryndon Museum. He added to the Museum a Centre for Prehistory and Palaeontology by attracting research grants from a number of sources, notably the Wenner-Gren Foundation for Anthropological Research and the National Geographic Foundation. By now the tide of his discoveries was beginning to flood; sites such as Rusinga, Olorgesailie and Olduvai producing important new finds.

The recovery of the 1948 "*Proconsul africanus*" skull was of great significance as it was the earliest ape skull then known. It was examined by Leakey and Le Gros Clark along with a great deal more material recovered from the Kenyan Miocene. The resulting monograph *The Miocene Hominoidea of East Africa* is still a foundation reference for those embarking upon primate palaeontology. The particular importance of the 1948 "*Proconsul africanus*" skull was that at that time it was considered to be the common ancestor of both modern apes and modern man. Subsequent finds made by Leakey, and work by others now make this unlikely. Leakey remained convinced, however, that the common ancestor of man and the modern apes would be found in Africa. The discovery of the Olorgesailie tool site was a major triumph as it remains one of the most significant collections of palaeolithic stone tools ever found.

Soon after the war Leakey was the driving force behind the first Pan-African congress on Prehistory and Quaternary Studies. The first meeting was held in Nairobi in 1947; it has continued at intervals of four years and has proved to be a valuable link

between prehistorians, geologists and anthropologists working in Africa. The name of Leakey will always be associated with Olduvai Gorge not only because of Louis but also because of his wife, Mary, who still works at the site. It was from Olduvai that his major success came, aided as always by his family. In 1959 Mary Leakey found the skull of "*Zinjanthropus*" (*Australopithecus boisei*) in Bed I. The skull is a magnificent example of the larger form of australopithecine; the first of its kind from East Africa and reliably dated at 1.75 m.yr. BP. Numerous other finds came from Olduvai including the remains of the smaller *Homo habilis* and from a higher level an example of *Homo erectus*, the maker of an early handaxe culture at Olduvai. At Fort Ternan excavations in Late Miocene deposits produced the maxilla of "*Kenyanthropus wickeri*"; still one of the earliest known hominid fragments but now usually accepted as *Ramapithecus wickeri*.

In scientific circles Leakey was always a controversial figure in a field renowned for controversy; he was an eager contestant in verbal or written debate and a keen participant at international meetings. He was the author of over twenty books and 120 scientific papers. Recognition came to him from the worldwide scientific community; he was awarded the Swedish Andre medal, the Cuthbert Peek medal of the Royal Geographic Society, the Viking medal of the Wenner-Gren Foundation for Anthropological Research, the Pitt-Rivers memorial medal of the Royal Anthropological Institute and, with his wife, Mary, the Prestwich medal of the Geological Society of London (the first time that this medal has been jointly awarded to a husband and wife team).

Only the perception of history will allow a true evaluation of Leakey's place in science since many of his opinions have yet to be vindicated or disproved. In the meantime we can only admire his enthusiasm, his initiative, his diligence and his consistent success in the search for fossils and stone tools. A man of greatness and courage who fought hard for the new knowledge that he gave to the world; a man who gave unstintingly of himself to the study of palaeontology, prehistory and the evolution of man.

Announcements

Miscellaneous

THE following have been awarded Alfred P. Sloan Research Fellowships. **Arizona State University:** Alexandra Navrotsky. **California Institute of Technology:** Michael Aschbacher, Jeffrey E. Mandula, Robert W. Vaughan, Michael W. Werner. **Stanford University:** John H. Coates, Theodor W. Hansch. **University of California, Berkeley:** Carl E. Heiles, Wayne L. Hubbell. **University of California, Irvine:** Robert T. McIver, Jr. **University of California, Los Angeles:** Thomas M. Liggett, Malcolm F. Nicol, Bruce L. Rothschild. **University of California, San Diego:** Carl H. Fitzgerald. **University of California, Santa Barbara:** David S. Cannell. **University of Colorado:** Arlan D. Norman. **Yale University:** Ronnie Lee, Bernard Shiffman. **Northwestern University, Evanston:** George Gasper, Jr. **Southern Illinois University, Carbondale:** Conrad C. Hinckley. **University of Chicago:** Robert D. Carlitz, Ronald H. Kluger, Paul B. Moore, Anthony D. J. Robertson. **University of Illinois, Urbana:** E. Graham-Evans, Jr., J. Douglas McDonald. **Iowa State University:** Jon C. Clardy. **University of Kentucky:** Thomas A. Chapman. **University of Maryland:** David D. Brayshaw. **Brandeis University:** David Eisenbud. **Harvard Medical School:** John G. Hildebrand. **Harvard University:** Bryan E. Kohler. **Massachusetts Institute of Technology:** Carleton DeTar, Michael S. Feld, Norberto Kerzman, K. Barry Sharpless. **Michigan State University, East Lansing:** Stanley R. Crouch, Richard T. Miller. **University of Michigan, Ann Arbor:** Hans H. Brintzinger. **University of Minnesota:** Ernest Coleman, Donald G. Truhlar. **University of Washington, Missouri:** Alfred G.

Hortmann. **Princeton University:** Edward J. Groth, III. **Columbia University:** C. Herbert Clemens, Paul G. Richards. **Cornell University:** Stuart J. Edelstein, Martin F. Semmelhack. **New York University:** Mark A. Ratner. **Rockefeller University:** Anthony Zee. **State University of New York at Stony Brook:** Philip B. Allen, Barry M. McCoy. **University of Rochester:** Stephen L. Olsen. **Duke University:** Thomas T. Norton. **University of North Carolina:** Reuben D. Rieke. **Case Western Reserve University:** Robert C. Dunbar, Raymond J. Lasek. **Reed College, Oregon:** Stephen W. Arch. **University of Pittsburgh:** David Jasnow, Ralph Z. Roskies. **Brown University:** James T. McIlwain, Langley A. Spurlock, Terry E. Tullis. **Vanderbilt University:** William B. Peatman. **Rice University:** W. Edward Billups. **University of Texas:** Kenneth E. Gentle, Jack B. Swift, Robert E. Wyatt. **University of Utah:** John P. Simons, Chin-Hsien Wang. **University of Virginia:** W. Lester S. Andrews. **University of Washington, Seattle:** John M. Wallace. **West Virginia University:** Mohindar S. Sehra. **University of Wisconsin:** Gerald L. Hazelbauer, Max G. Lagally, John L. Schrag. **Queen's University, Kingston, Ontario:** Ian P. Johnstone. **University of British Columbia, Vancouver:** Walter N. Hardy. **University of Waterloo, Ontario:** F. R. McCourt. **University of Windsor, Ontario:** Gordon W. F. Drake.

Errata

IN the article "Stereoselectivity and Differential Metabolism *in vivo* of Dextro and Laevo-Methadone-1-³H" by A. J. Misra and S. J. Mule (*Nature*, **241**, 281; 1973) the following corrections should be made:

Table 1, in the half-life column substitute 1.2 in place of 1.02; paragraph 2,

line 18, substitute 55.49 ± 2.15 (s.e.) for 57.3, and 29.77 ± 0.59 (s.e.) for 28.3. Legend for Fig. 3, line 13 "residues" should read "residue"; paragraph 2, line 32 should read "... differences in reversible plasma protein binding of two isomers ..."; paragraph 3, line 4, the reference above "l-isomer" should be deleted; paragraph 5, line 8, "d-methadone" should read "d-methadol"; paragraph 5, line 2, the reference above 'l-isomer' should be deleted.

IN the review of *Stabilité Structurale et Morphogénèse*, by René Thom (*Nature*, **242**, 207; 1973), the bibliography omitted to mention that Professor Thom's book is one of a series whose general editor is A. S. Wightman. The location of W. A. Benjamin was given as New York, instead of Reading, Massachusetts, which is the correct location.

International Meetings

June 2-8, **Première Semaine Mondiale de l'Innovation** (Inova 73, 62 Rue de Miro-mesnil, 75008 Paris, France).

June 4-7, **The 24th Annual Meeting of the Tissue Culture Association** (Local Committee Chairman, Dr George Yerganian, Children's Cancer Research Foundation, 35 Binney Street, Boston, Massachusetts 02115).

June 5-6, **The Quality of Life** (The Secretary, Institute of Rural Life at Home and Overseas, 27 Northumberland Road, New Barnet, Hampshire).

June 6-8, **Mode of Antibiotics on Microbial Walls and Membranes** (The Associate Executive Director, The New York Academy of Sciences, 2 East 63rd Street, New York, NY 10021 USA).

June 11-13, **Recent Advances in Staphylococcal Research** (The Associate Executive Director, The New York Academy of Sciences, 2 East 63rd Street, New York, NY 10021, USA).

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